TOXICOLOGICAL REVIEW

OF

HEXAVALENT CHROMIUM

(CAS No. 18540-29-9)

In Support of Summary Information on the Integrated Risk Information System (IRIS)

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FOREWORD

The purpose of this Toxological Review is to provide scientific support and rationale for the hazard identification and dose-response assessment in IRIS pertaining to chronic exposure to Cr(VI). It is not intended to be a comprehensive treatise on the chemical or toxicological nature of Cr(VI).

In Section 6, EPA has characterized its overall confidence in the quantitative and qualitative aspects of hazard and dose response. Matters considered in this characterization include knowledge gaps, uncertainties, quality of data, and scientific controversies. This characterization is presented in an effort to make apparent the limitations of the assessment and to aid and guide the risk assessor in the ensuing steps of the risk assessment process.

For other general information about this assessment or other questions relating to IRIS, the reader is referred to EPA's Risk Information Hotline at 513-569-7254.

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This document and summary information on IRIS have received peer review both by EPA scientists and by independent scientists external to EPA. Subsequent to external review and incorporation of comments, this assessment has undergone an Agencywide review process whereby the IRIS Program Director has achieved a consensus approval among the Office of Research and Development; Office of Air and Radiation; Office of Prevention, Pesticides, and Toxic Substances; Office of Solid Waste and Emergency Response; Office of Water; Office of Planning and Evaluation; and Regional Offices.

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Summaries of the external peer reviewers' comments and the disposition of their recommendations are in Appendix A.

LIST OF ABBREVIATIONS

BAL	Bronchoalveolar lavage
BALF	Bronchoalveolar lavage fluid
BCF	Bioconcentration factor
BMC	Benchmark concentration
BMD	Benchmark dose
CASRN	Chemical Abstracts Service Registry Number
CS	Composite score
ESADDI	Estimated safe and adequate dietary intake
IRIS	Integrated Risk Information System
LDH	Lactate dehydrogenase
LOAEL	Lowest-observed-adverse-effect level
LOEL	Lowest-observed-effect level
MCH	Mean corpuscular hemoglobin
MCV	Mean corpuscular volume
MED	Minimum effective dose
NOAEL	No-observed-adverse-effect level
NOEL	No-observed-effect level
PCMR	Proportionate cancer mortality ratio
ppm	Parts per million
RfC	Inhalation reference concentration
RfD	Oral reference dose
Rvd	Dose-rating value
Rve	Effect-rating value
SIR	Standard incidence ratio
SMR	Standard mortality ratio
TWA	Time-weighted average

1. INTRODUCTION

This document presents background and justification for the hazard and dose-response assessment summaries in EPA's Integrated Risk Information System (IRIS). IRIS Summaries may include an oral reference dose (RfD), inhalation reference concentration (RfC) and a carcinogenicity assessment.

The RfD and RfC provide quantitative information for noncancer dose-response assessments. The RfD is based on the assumption that thresholds exist for certain toxic effects such as cellular necrosis but may not exist for other toxic effects such as some carcinogenic responses. It is expressed in units of mg/kg-day. In general, the RfD is an estimate (with uncertainty spanning perhaps an order of magnitude) of a daily exposure to the human population (including sensitive subgroups) that is likely to be without an appreciable risk of deleterious effects during a lifetime. The inhalation RfC is analogous to the oral RfD. The inhalation RfC considers toxic effects for both the respiratory system (portal-of-entry) and for effects peripheral to the respiratory system (extrarespiratory or systemic effects). It is generally expressed in units of mg/m^3.

The carcinogenicity assessment provides information on the carcinogenic hazard potential of the substance in question and quantitative estimates of risk from oral exposure and inhalation exposure. The information includes a weight-of-evidence judgment of the likelihood that the agent is a human carcinogen and the conditions under which the carcinogenic effects may be expressed. Quantitative risk estimates are presented in three ways. The *slope factor* is the result of application of a low-dose extrapolation procedure and is presented as the risk per mg/kg-day. The *unit risk* is the quantitative estimate in terms of either risk per µg/L drinking water or risk per $\mu g/m^3$ air breathed. Another form in which risk is presented is a drinking water or air concentration providing cancer risks of 1 in 10,000; 1 in 100,000; or 1 in 1,000,000.

Development of these hazard identifications and dose-response assessments for hexavalent chromium has followed the general guidelines for risk assessment as set forth by the National Research Council (1983). EPA guidelines that were used in the development of this assessment may include the following: the *Guidelines for Carcinogen Risk Assessment* (U.S. EPA, 1986a), *Guidelines for the Health Risk Assessment of Chemical Mixtures* (U.S. EPA, 1986b), *Guidelines for Mutagenicity Risk Assessment* (U.S. EPA, 1986c), *Guidelines for Developmental Toxicity Risk Assessment* (U.S. EPA, 1991), *Proposed Guidelines for Neurotoxicity Risk Assessment* (U.S. EPA, 1995a), *Proposed Guidelines for Carcinogen Risk Assessment* (U.S. EPA, 1996a), *Guidelines for Reproductive Toxicity Risk Assessment* (U.S. EPA, 1996b), and *Guidelines for Neurotoxicity Risk Assessment* (U.S. EPA, 1998a); *Recommendations for and Documentation of Biological Values for Use in Risk Assessment* (U.S. EPA, 1988); (proposed) *Interim Policy for Particle Size and Limit Concentration Issues in Inhalation Toxicity* (U.S. EPA, 1994a); *Methods for Derivation of Inhalation Reference Concentrations and Application of Inhalation Dosimetry* (U.S. EPA, 1994b); *Peer Review and Peer Involvement at the U.S. Environmental Protection Agency* (U.S. EPA, 1994c); *Use of the Benchmark Dose Approach in Health Risk Assessment* (U.S. EPA, 1995b); *Science Policy Council Handbook: Peer Review* (U.S. EPA, 1998b); and memorandum from EPA Administrator, Carol Browner, dated March 21, 1995, Subject: Guidance on Risk Characterization.

Literature search strategies employed for this compound were based on the CASRN and at least one common name. At a minimum, the following databases were searched: RTECS, HSDB, TSCATS, CCRIS, GENETOX, EMIC, EMICBACK, DART, ETICBACK, TOXLINE, CANCERLINE, MEDLINE AND MEDLINE backfiles. Any pertinent scientific information submitted by the public to the IRIS Submission Desk was also considered in the development of this document.

2. CHEMICAL AND PHYSICAL INFORMATION RELEVANT TO ASSESSMENTS

In the hexavalent state, chromium exists as oxo species such as CrO_3 and CrO_4^{2-}) that are strongly oxidizing (Cotton and Wilkinson, 1980). The CAS Registry numbers and the solubilities of a few important hexavalent chromium compounds are given in Table 1.

In solution, hexavalent chromium exists as hydrochromate ($HCrO_4^-$), chromate (CrO_4^{2-}), and dichromate ($Cr_2O_7^{2-}$) ionic species. The proportion of each ion in solution is pH dependent. In basic and neutral pH, the chromate form predominates. As the pH is lowered (6.0 to 6.2), the hydrochromate concentration increases. At very low pH, the dichromate species predominate (U.S. EPA, 1984).

The primary sources of hexavalent chromium in the atmosphere are chromate chemicals used as rust inhibitors in cooling towers and emitted as mists, particulate matter emitted during manufacture and use of metal chromates, and chromic acid mist from the plating industry (ATSDR, 1993). Hexavalent chromium in air eventually reacts with dust particles or other pollutants to form trivalent chromium (NAS, 1974); however, the exact nature of such atmospheric reactions has not been studied extensively. Both hexavalent and trivalent chromium are removed from air by atmospheric fallout and precipitation (Fishbein, 1981). The atmospheric half-life for the physical removal mechanism is dependent on the particle size and particle density. Chromium particles of small aerodynamic diameter (< 10 µm) will remain airborne for a long period (U.S. EPA, 1984).

Hexavalent chromium may exist in aquatic media as water-soluble complex anions and may persist in water. Hexavalent chromium is a strong oxidizing agent and may react with organic matter or other reducing agents to form trivalent chromium. The trivalent chromium will eventually be precipitated as $Cr_2O_3 \cdot xH_2O$. Therefore, in surface water rich in organic content, hexavalent chromium will exhibit a much shorter lifetime (Callahan et al., 1979).

Any hexavalent chromium in soil is expected to be reduced to trivalent chromium by organic matter. The primary processes by which the converted trivalent chromium is lost from soil are aerial transport through aerosol formation and surface water transport through runoff (U.S. EPA, 1984). Very little chromium is leached from soil because it is present as insoluble $Cr_2O_3 \cdot xH_2O$ (Fishbein, 1981).

The bioconcentration factor (BCF) for hexavalent chromium in fish muscle appears to be < 1.0 (U.S. EPA, 1980), but values of 125 and 192 were obtained for oyster and blue mussel, respectively (U.S. EPA, 1980).

Table 1. CAS numbers and aqueous solubilities of selected hexavalent chromium compounds

Compound	CAS No.	Water solubility
Ammonium chromate $(NH_4)_2CrO_4$	7788-98-9	40.5 g/100 mL at 30°C
Calcium chromate $CaCrO_4$	13765-19-0	2.23 g/100 mL at 20°C
Chromic acid CrO_3	1333-82-0	61.7 g/100 mL at 0°C
Potassium chromate K_2CrO_4	7789-50-6	62.9 g/100 mL at 20°C
Potassium dichromate K_2Cr2O_7	7789-50-9	4.9 g/100 mL at 0°C
Sodium chromate Na_2CrO_4	7775-11-3	87.3 g/100 mL at 30°C
Sodium dichromate dihydrate	7789-12-0	230 g/100 mL at 0°C

Sources: Weast, 1980; Hartford, 1979.

3. TOXICOKINETICS RELEVANT TO ASSESSMENTS

3.1. ABSORPTION FACTORS IN HUMANS AND EXPERIMENTAL ANIMALS

3.1.1. Oral

Gastrointestinal absorption of Cr(VI) occurs with greater efficiency than absorption of Cr(III), though absorption of ingested hexavalent chromium is estimated to be less than 5%. Donaldson and Barreras (1966) fed $Na^{51}CrO_4$ to rats and humans. Based on mean urinary excretion, ^{51}Cr, absorption was estimated to be 2.1% in humans. In rats, approximately 2% of the administered dose was absorbed based on fecal excretion of ^{51}Cr when $Na^{51}CrO_4$ was administered intragastrically. Intestinal absorption of hexavalent chromium appears to be significantly affected by contact with gastric juices. When $Na^{51}CrO_4$ was introduced to humans intraduodenally (avoiding contact with gastric juices), approximately half of the chromium was absorbed based on fecal excretion. Similar results were observed following intrajejunal administration in rats. Incubation of hexavalent chromium with gastric juices prior to intraduodenal or intrajejunal administration in humans and rats, respectively, virtually eliminated absorption of chromium. Absorption of trivalent chromium ($^{51}CrCl_3$) was not increased by intraduodenal or intrajejunal administration. The authors demonstrated that hexavalent chromium is reduced to the trivalent form by incubation with gastric juices and concluded that reduction of hexavalent chromium to the trivalent form in the stomach significantly reduces absorption by the oral route of exposure. Gastric juices have peak reductive capacity 2 to 4 hours after a meal and are at a minimum between meals and at night (DeFlora et al., 1987). Anderson et al. (1983) confirmed the low absorption of trivalent chromium in humans following oral administration of 200 µg of $CrCl_3$.

MacKenzie et al. (1959) administered $Na^{51}CrO_4$ to rats by gavage. Based on urinary excretion, absorption was estimated to be 6% in fasted rats and 3% in nonfasted rats. The rate of absorption was twofold greater than that observed following administration of $^{51}CrCl_3$ by gavage. Absorption of hexavalent chromium was found to be increased by three- to fivefold following intestinal administration of $Na^{51}CrO_4$, consistent with reduction of hexavalent chromium during passage through the stomach. Absorption of hexavalent and trivalent chromium was found to be less than 1.4% in the hamster (Henderson et al., 1979), while Visek (1953) estimated that less than 0.5% of ingested $CrCl_3$ was absorbed through the gastrointestinal tract of the rat.

3.1.2. Inhalation

A number of factors can influence the absorption of chromium following inhalation, including the size, oxidation state, and solubility of the chromium particles; the activity of alveolar macrophages; and the interaction of chromium with biomolecules following deposition in the lung (ATSDR, 1993). Absorption of inhaled chromium following occupational exposure has been demonstrated by measurement of chromium in the serum, urine, and hair of workers in the chromium industry (Minoia and Cavalleri, 1988; Randall and Gibson, 1987; Tossavainen et al., 1980).

Langard et al. (1978) demonstrated that water-soluble hexavalent chromium, such as chromic acid, is absorbed rapidly following inhalation by rats. However, insoluble hexavalent chromium, such as zinc chromate, is less well absorbed. Animals were exposed to zinc chromate dust at a level of 7.35 mg/m^3. After 0, 100, 250, and 350 min of exposure, the concentrations of chromium in the blood (µg/ml) were 0.007, 0.024, 0.22, and 0.31, respectively. In the second part of this study, rats were exposed to the same level for 6 hours on 4 consecutive days. Blood concentrations appeared to peak at the end of the second exposure and then began to decline slowly. Mean blood chromium values measured at the end of each exposure period averaged 0.03, 0.56, 0.46, and 0.34 µg/ml for exposures l-4, respectively. No significant differences in absorption as reflected by blood chromium levels were noted between the sexes or between day and night exposures.

Suzuki et al. (1984) exposed rats to potassium dichromate (VI) or Cr(III) trichloride by inhalation and determined that while lung clearance of both valence states was dependent on particle size, Cr(VI) was absorbed with threefold greater efficiency than Cr(III).

3.1.3. Metabolism

In vivo reduction of Cr(VI) to Cr(III) has been widely studied, and some characterizations can be made. Ingested hexavalent chromium is efficiently reduced to the trivalent form by the gastric juices (DeFlora et al., 1987). Hexavalent chromium can also be reduced to the trivalent form in the epithelial lining fluid of the lungs by ascorbate and glutathione. The reduction by ascorbate is more rapid than by glutathione, and results in a shorter residence time for chromium in the lungs (Suzuki and Fukuda, 1990).

Once absorbed into the bloodstream, hexavalent chromium readily enters red blood cells through the phosphate and sulfate anion-exchange carrier pathway, though a portion may remain in plasma for an extended period (Wiegand et al., 1985). While Cr(III) compounds are unable to cross the red cell membrane by this pathway (Gray and Sterling, 1950), they may enter red blood cells, but only with very low efficiency (Mertz, 1969; O'Flaherty, 1996). Hexavalent chromium is reduced to the trivalent form in the red blood cell by the action of glutathione (Debetto and Luciani, 1988; Petrilli and De Flora, 1978a). During reduction to the trivalent form, chromium may interact with cellular macromolecules, including DNA (Wiegand et al., 1985), or may be slowly released from the cell (Bishop and Surgenor, 1964).

Visek et al. (1953) reported that an insignificant amount of ^{51}Cr crossed the placenta of rats in the 24 hours following intravenous injection regardless of the chemical form injected, the valence state, the gestational stage, or the size of the litter. In no instance was the radioactivity measured in the fetuses greater than 0.13% of the dose. Danielsson et al. (1982) reported that sodium dichromate crossed the placenta more readily than Cr(III) trichloride following intravenous injection of mouse dams. Casey and Hembridge (1984) demonstrated that chromium can be transferred to infants through breast milk. The breast milk of 45 lactating women was found to have a chromium content averaging 0.3 µg/L. These concentrations were taken to represent background levels in women whose chromium exposure occurs primarily through the diet.

A physiologically based model for chromium has recently been developed, which incorporates absorption and disposition schemes for Cr(VI) and Cr(III) throughout the body (O'Flaherty, 1996). The model was calibrated on the basis of published oral and intratracheal kinetic studies using soluble Cr(III) and Cr(VI) in the rat, and accounts for most of the major features of chromium kinetics, including reduction of Cr(VI) to Cr(III). The model suggests the following in vivo disposition for chromium. Both Cr(III) and Cr(VI) are poorly absorbed from the lung and the gastrointestinal tract. Following inhalation exposure, chromium may be absorbed into the systemic circulation, transferred to the gastrointestinal tract by mucociliary action, or remain in the lung. Cr(VI) is reduced to Cr(III) in all tissues, including the lung and the gastrointestinal tract. Both Cr(III) and Cr(VI) are better absorbed from the gastrointestinal tract in the fasted than in the fed state, and the absorption efficiency of Cr(III) salts is largely dependent on the nutritional status of the animal as well as the nature of the anion making up the Cr(III) salt. The model assumes that reduction of Cr(VI) does not occur in the plasma. Cr(VI) enters cells by the phosphate and sulfate anion-exchange carrier pathway. Cr(III) travels in the bloodstream largely bound to amino acids, other organic acids, and plasma proteins such as globulins. The complexes of Cr(III) that are bound to lower molecular weight ligands are most likely to be able to traverse cell membranes (Mertz, 1969). A significant amount of absorbed chromium is taken up in the bone (Witmer and Harris, 1991; Weber, 1983), Chromium is also concentrated in tissues of the liver, kidney, and spleen. Once in the cell, Cr(VI) may be reduced to Cr(III), which may subsequently interact with cellular macromolecules including DNA (Wiegand et al., 1985), or may be slowly released from the cell (Bishop and Surgenor, 1964). The model suggests that the bioaccessibility of chromium to absorption processes may be the single most important factor determining the toxicity of a specific chromium source (O'Flaherty, 1996).

Given the rapid reduction of Cr(VI) to Cr(III) in vivo, it is relevant to consider whether environmental exposures to Cr(VI) or administration of Cr(VI) in controlled animal experiments is essentially identical to environmental exposures to Cr(III) or administration of Cr(III) in controlled experiments. Although considerably more data are available for Cr(VI) than for Cr(III), it appears at present that exposures to Cr(VI) have considerably different outcomes than exposures to Cr(III). The Agency has prepared the toxicological summaries and IRIS files for Cr(VI) and Cr(III) from this perspective.

3.1.4. The Essentiality of Chromium

Cr(III) in its biologically active form (glucose tolerance factor, or GTF, a dinicotinato-chromium[III] glutathione-like complex), facilitates interaction of insulin with its receptor site, influencing glucose, protein, and lipid metabolism. Thus, Cr(III) is essential for animals and human beings. Inorganic chromium compounds do not appear to have insulin-potentiating activity. Chromium deficiency may cause changes in the metabolism of glucose and lipids. In some studies, dietary supplementation with chromium reversed changes in glucose tolerance and serum lipids. The National Research Council has identified an estimated safe and adequate daily dietary intake (ESADDI) for chromium of 50-200 µg/d (NRC, 1989), corresponding to 0.71-2.9 µg/kg/day for a 70-kg adult. FDA has selected a Reference Daily Intake for chromium of 120 µg/d (DHHS, 1995).

4. HAZARD IDENTIFICATION

4.1. STUDIES IN HUMANS

4.1.1. Oral

Cr(VI) is considerably more toxic than Cr(III). A cross-sectional study of 155 villagers reported the effects of environmental contamination of well water adjacent to a chromium alloy plant. Cr(VI) concentrations were reported as 20 mg/L, with an estimated dose rate of 0.57 mg/kg-day (Zhang and Li, 1987). Reported effects at this dose included oral ulcers, diarrhea, abdominal pain, indigestion, vomiting, leukocytosis, and presence of immature neutrophils. Effects data for lower exposure doses were not available.

Other reports of toxic effects in humans are limited to case reports from accidental poisonings. Some Cr(VI) compounds (such as potassium tetrachromate and chromic acid) are potent oxidizing agents, and are thus strong irritants of mucosal tissue. Effects included metabolic acidosis, acute tubular necrosis, kidney failure, and death (Saryan and Reedy, 1988).

4.1.2. Inhalation

Occupational exposure to chromium compounds has been studied in the chromate-production, chrome-plating and chrome pigment, ferrochromium production, gold mining, leather tanning, and chrome alloy production industries.

Workers in the chromate industry are exposed to both trivalent and hexavalent compounds of chromium. Epidemiological studies of chromate production plants in Japan, Great Britain, West Germany, and the United States have revealed a correlation between occupational exposure to chromium and lung cancer, but the specific form of chromium responsible for the induction of cancer was not identified (Machle and Gregorius, 1948; Baejter, 1950a,b; Bidstrup, 1951; Mancuso and Hueper, 1951; Brinton et al., 1952; Bidstrup and Case, 1956; Todd, 1962; Taylor, 1966; Enterline, 1974; Mancuso, 1975; Ohsaki et al., 1978; Sano and Mitohara, 1978; Hayes et al., 1979; Hill and Ferguson, 1979; Alderson et al., 1981; Haguenor et al., 1981; Satoh et al., 1981; Korallus et al., 1982; Frentzel-Beyme, 1983; Langard and Vigander, 1983; Watanabe and Fukuchi, 1984; Davies, 1984; Mancuso, 1997).

Mancuso and Hueper (1951) conducted a proportional mortality study of a cohort of chromate workers (employed for > l year from 1931 to 1949 in a Painesville, OH, chromate plant) in order to investigate lung cancer associated with chromate production. Of the 2,931 deaths of males in the county where the plant is located, 34 (1.2%) were due to respiratory cancer. Of the 33 deaths among the chromate workers, however, 6 (18.2%) were due to respiratory cancer. Within the limitations of the study design, this report strongly suggested an increased incidence of respiratory cancer in the chromate-production plant.

In an update of the Mancuso and Hueper (1951) study, Mancuso (1975) followed 332 of the workers employed from 1931-1951 until 1974. By 1974, > 50% of this cohort had died. Of

these men, 63.6%, 62.5%, and 58.3% of the cancer deaths for men employed in 1931-1932, 1933-1934 and 1935-1937, respectively, were due to lung cancer. Lung cancer death rates increased with increases in exposure to total chromium, and significant deposition of chromium was found in the lungs of workers long after the exposure ceased. The age-specific lung cancer rates from and gradient exposures to total chromium from this study are presented in Table 2. Mancuso (1975) reported that these lung cancer deaths were related to insoluble (trivalent), soluble (hexavalent), and total chromium exposure, but the small numbers involved make identification of the specific form of chromium responsible for the lung cancer uncertain.

Mancuso (1997) recently updated this study, following the combined cohort of 332 workers until 1993. Of 283 deaths (85% of the cohort identified), 66 lung cancers were found (23.3% of all deaths and 64.7% of all cancers). Lung cancer rates clearly increased by gradient level of exposure to total chromium. The relationship between gradient level of exposure and lung cancer rates is less clear for trivalent and hexavalent chromium. The rates of lung cancer within the cohort are consistent with those reported in Mancuso (1975) and provide further support for the cancer risk assessment based on those data.

Hayes et al. (1979) conducted a study of 2,101 male workers in a chromium chemicals production plant in Baltimore, MD, to determine whether employment in a modernized facility resulted in a reduction in the risk of lung cancer in comparison with that seen in workers employed in older production facilities. Mortality data for the workers were compared with cause-specific mortality rates for Baltimore city males, and correlations were made between a history of having worked in specific job positions and cancer of the lung. Workers with lung cancer as an underlying or contributory cause of death were individually matched to controls selected from plant workers who died of causes other than cancer. Many of the specific causes of death, including several sites of malignancy, showed results well below the expected number. While the newer facility had been constructed to reduce exposure to carcinogens, workers in the new facility were still found to have an increased risk of lung cancer. Both short-term and long-term workers were found to have excess risk for lung cancer, but the effect was more pronounced in the long-term workers. A dose-response relationship with respect to length of employment was observed, and long-term laborers were found to have a maximum risk (standard mortality ratio [SMR] = 3.9) between 15 and 19 years after the initial exposure. A history of having worked with highly soluble hexavalent chromium compounds in the “wet end” of the production process was particularly associated with an increased risk of lung cancer. Among the workers in the low- exposure group (n = 699), no excess risk was demonstrated. The authors were unable to obtain suitable data to account for cigarette smoking in the study population, and assumed that the smoking habits of workers were similar to those of the in the general population.

Pastides et al. (1994) conducted a retrospective cohort study in the United States’ largest chromate chemicals manufacturing facility in Castle Hayne, NC. This plant was designed to reduce the high level of chromium exposure found at most of the older chromate production facilities. The study utilized more than 5,000 personal breathing zone samples collected over a 15-year period. Occupational, medical, and smoking data from current and former employees were collected using a questionnaire. A healthy-worker effect was observed in the population. There was no increased risk of mortality or cancer among employees who worked only at this

Table 2. Age-specific lung cancer death rates and gradient exposures to total chromium

Mg/m^3 - Yrs. total insoluble chromium								
Age		**< 1.00**	**1.0-1.99**	**2.0-3.99**	**4.0-5.99**	**6.0-6.99**	**7.0-7.99**	**8+[a]**
45-54	Deaths	1	2	2	4	3	3	0
	Person-years	886	459	583	348	159	140	262
55-64	Deaths	1	3	1	4	2	3	1
	Person-years	707	356	462	250	113	98	203
65-74	Deaths	1	1	2	1	1	0	3
	Person-years	235	166	182	80	42	41	81

[a]Data in the last column are not used in EPA's risk assessment because the range of exposure in this class is not known, and it does not appear reasonable to assume that all three age groups had an identical exposure distribution.

Source: Mancuso, 1975.

facility, though a subgroup of workers who transferred from older facilities was found to have higher risks of mortality and cancer.

Rosenman and Stanbury (1996) recently reported on a study of lung cancer risk in a population of former workers from four facilities that produced chromium compounds from chromite ore. Workers at these facilities between 1937 and 1971 were found to have proportionate cancer mortality ratios (PCMR) of 1.51 for white men and 1.34 for black men. The PCMRs were found to increase with duration of employment and time since first employment. The authors also reported a PCMR for nasal/sinus cancer of 5.18. The lung cancer risk remained elevated for more than 20 years following the cessation of exposure. While the study was unable to account for smoking habits of the workers, the lack of increase in other smoking-related diseases suggested that the lung cancer was not related to smoking. The authors were unable to distinguish risk from trivalent and hexavalent chromium, and were also unable to examine risk by exposure estimates.

Alexander et al. (1996) reported on a study of lung cancer in chromate-exposed aerospace workers with a minimum of 6 mo exposure to hexavalent chromium. Standard incidence ratios (SIRs) for lung cancer were determined on the basis of the duration of employment in chromate-exposure jobs and cumulative exposure. The authors reported an inverse relationship between lung cancer risk and estimates of cumulative chromate exposure in spray painters. Elevated lung cancer risks were found for subjects who worked as chrome platers or surface processor tank tenders, sanders/maskers, and polishers, but the numbers are too low to establish a clear association between chromate exposure and lung cancer risk in these workers. The authors suggested that the lack of increased lung cancer risk among spray painters may reflect the exposure to chromate in paint mists, which they suggested may render the hexavalent chromium less biologically active.

Studies of workers in chromate pigment production plants have demonstrated an association with increased risk of lung cancer. Hayes et al. (1989) studied workers employed between 1940 and 1969 in a lead and zinc chromate pigment production plant in New Jersey. The SMR for lung cancer was 160, based on comparison with U.S. lung cancer rates for white males. Workers in the high-exposure group were continuously exposed to greater than 2 mg/m^3 chromate dust; workers in the moderate-exposure group were occasionally exposed to concentrations between 0.5 and 2 mg/m^3 ; and workers in the low-exposure group were infrequently exposed at concentrations below 0.1 mg/m^3. Significant SMRs were also found for lung cancer in nonwhite males and stomach cancer in white males. Air monitoring revealed that exposures were lead chromate.

Davies (1978, 1979) studied three chromate pigment plants in England, two of which produced both zinc and lead chromate, and one which produced only lead chromate. The cohort of exposed workers consisted of employees with $\geq$1 year of service and for whom vital statistics were available as of 1977. Using these guidelines, 396 and 136 subjects were obtained from the plants producing both zinc and lead chromate, and 114 subjects were obtained from the plant that produced only lead chromate. The observed mortality from lung cancer in the different plants was compared to the expected mortality based on national lung cancer mortality rates for all

males in England and Wales. An elevated risk of lung cancer was present only in the plants producing both zinc and lead chromate. The authors suggested that these data indicated that zinc chromate was associated with the etiology of lung cancer, whereas lead chromate was not.

Davies (1984) conducted a follow-up study on workers in the same three chromate pigment production plants. Workroom chromium concentrations and smoking statistics for the workers were not available in any of the factories. Elevated SMRs for lung cancer were observed, particularly for workers employed prior to improvements in industrial hygiene. Workers in the high- and medium-exposure groups in a plant producing lead and zinc chromates had an SMR for lung cancer of 232. Workers in high- and medium-exposure groups in a second lead and zinc chromate production plant had a SMR for lung cancer of 373. The SMR for workers in high and medium groups exposed to lead, zinc, and strontium chromate was 562. In the third plant, only lead chromate pigments were produced, and no excess lung cancer deaths were reported.

Langard and Norseth (1975) reported on three pigment plants in Norway that were in operation between 1948 and 1972 (one of the plants was brought on line in the year the study ended). A total of 133 workers were identified as employees at the three plants during this time period. Of the 133, 24 had been employed > 3 years, and of this cohort, 3 cases of lung cancer were identified through the Cancer Registry of Norway. All of the lung cancer cases had been employed for 5 years or longer. Data from the cancer registry indicated an expected number of lung cancer cases among those employed of 0.079; thus, the observed number of cases was 38 times greater than expected, based on the general population. Exposure levels determined by personal monitoring were reported for the plants for the year 1972, with chromium levels in the two older plants ranging between 0.04 and 1.35 mg/m^3 and levels in the new plant between 0.01 and 0.08 mg/m^3. Although an increased risk of lung cancer was indicated, two of the individuals with lung cancer were moderate to heavy smokers. Nevertheless, a relative risk of lung cancer of 38 could not be explained by differences in smoking between the study cohort and the Norwegian population. Langard and Vigander (1983) conducted a follow-up study on workers employed for at least 3 years in the same chromate pigment producing factories. Workroom monitoring revealed significantly elevated concentrations of hexavalent chromium (0.01 - 1.35 mg/m^3), and an SMR for lung cancer of 4,444 was reported. Workers in this group were exposed over a period of 6-9 years.

Frentzel-Beyme (1983) reported that the observed number of lung cancer deaths exceeded those expected among workers in five chromate pigment plants in the Netherlands and West Germany. In only one factory, however, was this excess statistically significant. The authors did not find a lung cancer mortality dose-response by intensity or duration of exposure. The analysis was limited by the numbers of deaths in each category.

Several studies of the chrome-plating industry have demonstrated a positive relationship between cancer and exposure to chromium compounds (Royle, 1975; Franchini et al., 1983; Sorahan et al., 1987). Royle (1975) studied mortality in the chromium plating industry in England in a retrospective study between 1969 and 1972. Workers in this industry are exposed to hexavalent chromium in the form of chromic acid mist and some sodium dichromate dust.

The study traced 1,238 chrome plating workers employed for > 3 months along with 1,284 manual laborers used as controls. There was little difference in smoking habits between the groups. There was a significant ($p < 0.05$) difference in the death rate for cancer at all sites, 3.15% in chrome platers as compared to 1.63% in controls. Deaths from malignancy of the lung and gastrointestinal tract were each increased, though not significantly.

Franchini et al. (1983) conducted a retrospective cohort study of 178 workers in nine Italian chromeplating plants to determine the mortality of workers employed for at least 1 year between January 1951 and December 1981. The mortality experience of workers was compared to that for the Italian male population of the same age during the follow-up period. Direct exposure measures from the plants were not available, but the exposures were related to airborne chromium concentrations taken from Italian electroplating plants in 1980, after industrial hygiene practices had improved considerably. A significant excess of all malignancies and lung cancer specifically was found for the workers in "hard" chromeplating plants, who were expected to have the greatest chromium exposures. While the small size of the cohort limited the statistical power of the study and confounding factors were not assessed, this study is taken to provide suggestive evidence of a causal relationship between exposure to chromic acid and cancer.

Sorahan et al. (1987) conducted a mortality study of nickel/chromium platers in England who were employed between 1946 and 1983. Exposure in these the plants was to soluble hexavalent chromic acid mist (CrO_3). The cohort in the study included a population of 2,689 workers (1,288 men, 1,401 women). Workers' exposure to chromium was estimated based on participation in any of eight chromeplating job categories and on cumulative duration of employment in chrome bath jobs. The mortality experience of the cohort was evaluated through comparison with that expected for the general population of England and Wales, as well as through comparison of the estimated chrome exposures of workers who died in a given year with those of matching survivors in the same follow-up year, controlling for sex, year of starting chrome employment, and age starting in chrome employment. Overall, compared with the general population, significant differences were found for all cancers, cancers of the lung and bronchus, cancer of the nose and nasal cavities, cancer of the stomach, and primary cancer of the liver. The results were particularly striking for chrome bath workers, who were likely the most heavily exposed to chromium. Significant positive associations were found between cancers of the lung and bronchus and duration of chrome bath work.

Some studies of the chromeplating industry have reported inconclusive results (Silverstein et al., 1981; Okubo and Tsuchiya, 1979; Takahashi and Okubo, 1990; Itoh et al., 1996). Okubo and Tsuchiya (1979) conducted a cohort study of 889 Tokyo chrome platers, with an unspecified number of controls selected from the same factories. The study included a 6-year follow-up period. The investigation was conducted by a questionnaire sent to the manager of each factory, and vital statistics were ascertained using the records of the Tokyo Health Insurance Society of the Plating Industry. The recovery rate of the questionnaire was 70.5%. Among the 889 male chromium platers, 19 deaths were observed, or about 50% of those expected (healthy-worker effect). In contrast, the authors reported a slightly higher percentage of deaths in the control group. The authors reported negative results for the relationship between chromeplating

and lung cancer; however, the results were not related to well-defined exposure data and the study utilized a very short follow-up period.

Takahashi and Okubo (1990) reported on an epidemiological study of metal platers in 415 small chromeplating plants in Japan. Members of the cohort were all male workers employed as platers for at least 6 mo between April 1970 and September 1976. The follow-up period extended until 1987 and no members of the cohort were lost to follow-up. The members of the cohort were classified into two subgroups based on their work histories: 52% of the cohort had more than 6 mo experience in chrome plating and 48% of the cohort had more than 6 mo plating experience using metals other than chromium. The mean duration of exposure before the follow-up period was 12.3 years. The study lacked direct exposure measures, and smoking histories for the workers were not available. All-cause mortality in the cohort was slightly below the expected number (healthy-worker effect). The study demonstrated that exposure to metal plating is associated with a statistically significant increase in lung cancer, though the elevated SMR was not statistically significant in either of the two plater subgroups. While chromium platers with initial exposure prior to 1960 had a 2.5-fold (though statistically insignificant) excess incidence of lung cancer, there was no increase in numbers of deaths due to lung cancer relative to the length of time exposed to chromeplating. The results of this study are considered equivocal regarding the relationship between chromeplating and lung cancer. Itoh et al. (1996) subsequently reported on a prospective cohort study of the same cohort of 1,193 workers at small Japanese plating facilities. While a trend toward statistical significance for risk of lung cancer was seen in the chromium plating subgroup, the study lacked sufficient statistical power to form a clear conclusion.

Silverstein et al. (1981) found a statistically significant increase ($p < 0.001$) in the lung cancer proportionate mortality ratios for both male and female white employees in a die-casting and electroplating plant. In this plant, workers were exposed to chromium during electroplating, but nickel and copper were also used in electroplating. Other operations of the plant included zinc alloy die-casting, buffing and polishing, and cleaning of zinc and steel parts. No conclusion can be made from this study regarding the association of chromium electroplating and lung cancer mortality, because of the employees' exposure to other potential carcinogens.

A number of epidemiological studies have considered the association between inhalation of Cr(VI) and noncarcinogenic endpoints, including upper respiratory irritation and atrophy, lower respiratory effects, and systemic effects.

Bloomfield and Blum (1928) examined 23 men from 6 chromium plating plants in the United States. Fourteen of these workers typically spent 2-7 hours/day over vats of chromic acid, which generated airborne hexavalent chromium ranging from 0.12 to 5.6 mg/m^3. These men experienced nasal tissue damage, including perforated septum (2), ulcerated septum (3), chrome holes (6), nosebleed (9), and inflamed mucosa (9). In general, the nine remaining workers examined, not directly exposed to chromium vapors, had only inflamed mucosae. The authors concluded that chromic acid at concentrations greater than 0.1 mg/m^3 is likely to cause nasal tissue injury. However, no concentrations lower than 0.12 mg/m^3 were observed, and injury to nasal tissue caused by lower concentrations could not be ruled out.

Machle and Gregorius (1948) reported an incidence of nasal septal perforation of 43.5% in 354 employees who worked in a chromate-producing plant that manufactured sodium chromate and bichromate. At the time of the study, airborne chromate concentrations ranged from 10 to 2,800 $\mu g/m^3$. The plant had been in operation for at least 17 years, and some employees probably worked in the plant when reverberatory furnaces, a prominent source of high chromate exposure, were used.

Mancuso and Hueper (1951) reported on physical examinations of a random sample of 97 workers from a chromate-chemical plant. The results indicated that 61 of the 97 workers (63%) had septal perforation. The data suggested to the author that Cr(III) may be partly responsible for the perforations; however, there are insufficient data to make an unequivocal conclusion.

The U.S. Public Health Service conducted a study of workers in seven chromate-producing plants in the early 1950s. Of 897 chromate industry workers in the study, 57% were found to have a nasal septum perforation. Perforated septum was observed even in workers employed less than 6 mo The study indicated that exposure to chromate results in severe nasal tissue destruction, but exposure levels were not measured; hence, the data are of limited usefulness for risk assessment purposes (Federal Security Agency, 1953).

Vigliani and Zurlo (1955) reported nasal septal perforation in workers exposed to chromic acid and chromates in concentrations of 0.11-0.15 mg/m^3. The lengths of exposure were not known. Hanslian et al., (1967) reported on otolaryngologic examinations of 77 persons exposed to chromic acid aerosol during chrome plating. Among this group, 19% were observed to have septal perforation and 48% to have nasal mucosal irritation. The workers averaged 6.6 years of exposure to an airborne chromium concentration of 0.4 mg/m^3. In 14 persons, papillomas of the oral cavity and larynx were found. The diagnosis of papilloma was confirmed by histologic examination. There were no signs of atypical growth or malignant degeneration.

Kleinfeld and Russo (1965) reported some degree of nasal septal ulceration in 7 of 9 workers in a chromeplating plant, with 4 of 7 demonstrating frank perforations. Analyses of air samples showed chromium concentrations of 0.18-1.4 mg/m^3. Data regarding the length of exposure and exposure concentration for individual workers were not available.

Gomes (1972) examined 303 employees who worked in 81 electroplating operations in Sao Paulo, Brazil. More than two-thirds of the workers had mucous membrane or cutaneous lesions, with many of them having ulcerated or perforated nasal septa. The duration of exposure was not stated, but the author mentioned that the harmful effects were noted in < l year. A direct correlation between workers exposed to a given airborne concentration of Cr(VI) and the development of harmful effects could not be made.

Cohen and Kramkowski (1973) and Cohen et al. (1974) examined 37 workers (7 male and 30 female) employed in the nickel-chrome department of an electroplating plant in comparison with 21 workers (15 male and 6 female) in other areas of the plant not significantly exposed to chromic acid. Smoking demographic data were not provided. Environmental air samples were collected from breathing zones of several workers in the exposed and control

groups to determine concentrations of total chrome and Cr(VI). Brief medical histories were confined to the ear, nose, throat, and cutaneous structures. Within l year of employment, 12 workers experienced nasal ulceration or perforation. Nasal ulcers and perforations were associated with total chromium concentrations of 1.4 to 49.3 $\mu g/m^3$, averaging 7.1 $\mu g/m^3$, and Cr(VI) concentrations of 0.09 to 9.1 $\mu g/m^3$, averaging 2.9 $\mu g/m^3$. Ninety-five percent of the 37 workers studied exhibited pathologic changes in nasal mucosa in a concentration-duration response. More than half of the workers employed less than 1 year had nasal pathology that was more severe than simple redness of the nasal mucosa. Almost all the workers (35 of 37) employed longer than 1 year had nasal tissue damage. The authors noted the lack of good industrial hygiene practices, implicating direct contact, such as touching of the nose with chromium-contaminated hands, as a potentially important route of exposure.

Lucas and Kramkowski (1975) conducted a health hazard evaluation of 11 employees in the "hard" chrome area of an industrial plating facility. The average age of the employees was 39 years and the average duration of employment in the hard chrome area was 7.5 years. Medical examinations were conducted to evaluate the presence of dermatitis, chrome holes, old chrome hole scars, ulcerated nasal septum, infection of the mucosa, nasal redness, perforated nasal septum, reddened throat, conjunctivitis, and wheezing. Environmental air samples were collected from the breathing zone on all workers in the hard chrome area to determine the concentrations of hexavalent chromium. Cr(VI) concentrations ranged from 1 to 20 $\mu g/m^3$, averaging 4 $\mu g/m^3$. However, the authors attributed the nasal pathology primarily to direct contact. Clinical observations included injection of the nasal mucosa in five workers, ulcerated nasal septum in two workers, atrophic scarring indicative of the presence of past ulceration in two workers, and complete perforation of the nasal septum in four workers. Poor hygiene practices including touching the nose with the hand were noted at the plant and represented a confounding factor in the etiology of the nasal lesions.

Markel and Lucas (1973) conducted a health hazard evaluation of 32 workers at a "cold dip" chromeplating plant who were employed in the chrome department or who regularly spent a portion of their workday in that area. Twenty of the employees worked in the chrome area of the plant for more than 5 years. A total of 16 personal and 7 general air samples were taken to determine the concentrations of Cr(VI). Maximum airborne Cr(VI) concentration was 3 $\mu g/m^3$. No workers were found to have ulcerated nasal mucosa or perforated nasal septa. Half of the 32 employees had varying degrees of mucosal irritation. The authors did not consider this to be significant, because the survey was carried out at the peak of the 1972-1973 influenza epidemic.

Lindberg and Hedenstierna (1983) compared lung function, the condition of the nasal septum, and subjective symptoms related to respiratory health (data obtained by questionnaire) in unexposed controls (119) and workers (43) exposed to chromic acid in chromeplating operations. Exposed workers were divided into low- (< 2) and high- (> 2 μg Cr[VI]/m^3) exposure groups based on the exposures they were likely to have experienced in the workplace. Complaints of diffuse nasal symptoms ("constantly running nose," "stuffy nose," or "a lot to blow out") were registered by 4/19 workers in the low-exposure group and half of the 24 workers in the high exposure group. The authors reported reddening of the nasal mucosa at 1 to 2 $\mu g/m^3$ and nasal irritation (chronic and nasal septal ulceration and perforation) in two-thirds of the subjects at

concentrations from 2 to 20 $\mu g/m^3$. All workers with nasal ulceration had been exposed to chrome acid mist, which contained Cr(VI) at 20 $\mu g/m^3$, or greater than 20 $\mu g/m^3$ near the baths. Changes in pulmonary function measurements, as determined by changes in vital capacity and forced expiatory volume at 1 sec (FEV_1), were seen in workers who experienced Cr(VI) exposures greater than 2 $\mu g/m^3$. Examination of the nasal septum revealed that damage was significantly greater in exposed workers than in unexposed controls and appeared to be somewhat more severe in the high-exposure group than the low-exposure group. There was a tendency for lung function parameters to return to normal over a 2-day weekend.

In the United States, 97 workers in chromate-producing plants had a higher incidence of severely red throats and pneumonia, but did not show any increase in the incidence of other respiratory diseases when compared with control groups. Although bilateral hilar enlargement was observed, there was no evidence of excessive pulmonary fibrosis in these workers (Federal Security Agency, 1953). The various lung changes described in these workers may represent a nonspecific reaction to irritating material or a specific reaction to chromium compounds. Many of the conditions mentioned occur widely in the general population (NAS, 1974).

Lindberg and Vesterberg (1983a) studied urinary excretion of proteins in 24 currently employed chrome platers and 27 former chrome platers. Results were compared with those for a group of 37 referents. Exposures for current workers were determined using personal samplers and were found to range from 2 to 20 $\mu g/m^3$, with an average level of 6 $\mu g/m^3$. Exposure levels of former platers were thought to be higher than those for the current workers. The duration of exposure ranged from < 1 to 26 years. Cr(VI) exposure was found to result in renal effects in a dose-dependent fashion (based on elevated excretion of β-2-microglobulin as an indicator of nephrotoxicity) in current workers exposed to 4 to 20 $\mu g/m^3$ Cr(VI) over 8-hour shifts. The effect may be reversible because former chromeplaters did not have an elevated concentration of either β-2-microglobulin or albumin in their urine. Most of the currently exposed workers were also observed to have irritation symptoms of the airways, including ulcerated nasal septum and complete perforations. Severe objective and subjective levels for the airway effects occurred at NOAEL levels for renal toxicity.

In another study, Saner et al. (1984) did not find increased urinary β-2-microglobulin levels in tannery workers in comparison to referent control workers. However, comparison of urinary chromium concentrations of the tannery workers in this study versus the chromeplaters in the Lindberg and Vesterberg (1983a,b) study suggests that the latter had distinctly higher chromium exposures.

Various other disease states have been attributed to chromium, but in most cases, the etiologic relation to chromium is doubtful because of the presence of other chemicals (NAS, 1974). These studies, reviewed by EPA (1984) and ATSDR (1993), will not be reviewed here.

4.2. SUBCHRONIC, CHRONIC STUDIES AND CANCER BIOASSAYS IN ANIMALS

4.2.1. Chronic Oral Studies

Only one chronic study pertaining to the oral toxicity of hexavalent chromium was located in the available literature. Anwar et al. (1961) exposed dogs orally to potassium chromate in drinking water for 4 years. Treatment levels were 0, 0.45, 2.25, 4.5, 6.75, and 11.2 ppm potassium chromate; there were two dogs/group. No effects were observed with regard to gross and microscopic analysis of all major organs, urinalysis, and weights of spleen, liver, and kidney.

4.2.2. Subchronic Oral Studies

The National Toxicology Program recently conducted a three-part reproductive toxicity study to investigate oral ingestion of hexavalent chromium in experimental animals (NTP, 1996a,b, 1997). Rats and mice were exposed to 0 - 400 ppm potassium dichromate daily in the diet for 9 weeks. Animals were examined for body weights; feed and water consumption; organ weights; microscopic evaluation of the liver, kidney, and ovaries; hematology; histology of the testis and epididymus for Sertoli nuclei and preleptotene spermatocyte counts in Stage X or XI tubules; and chromatin analysis. No treatment-related hematology findings were reported except for slight decreases in mean corpuscular volume (MCV) and mean corpuscular hemoglobin (MCH) values in the male and female treatment groups receiving 400 ppm potassium dichromate (24 mg/kg-day). The findings were characterized by the authors as suggestive of a potential bone marrow/erythroid response. The authors considered the 100 ppm (6 mg/kg-day) dose group to be representative of the NOAEL for the study. The studies are described in greater detail in the reproductive/developmental section of this document.

MacKenzie et al. (1958) exposed groups of eight male and eight female Sprague-Dawley rats to 0-11 ppm (0-11 mg/L) hexavalent chromium (as K_2CrO_4) for 1 year in drinking water. The control group (10/sex) received distilled water. A second experiment involved three groups of 12 male and 9 female rats. One group was given 25 ppm (25 mg/L) chromium (as K_2CrO_4), a second group received 25 ppm chromium in the form of chromic chloride, and the controls again received distilled water. The results of the MacKenzie et al. study are presented in Table 3. No significant adverse effects were seen on appearance, weight gain, or food consumption, and there were no pathologic changes in the blood or other tissues in any treatment group. The rats receiving 25 ppm of chromium (as K_2CrO_4) showed an approximate 20% reduction in water consumption. This dose corresponds to 2.5 mg Cr(VI)/kg/day based on actual body weight and water consumption data. Blood was examined monthly, and tissues (livers, kidneys, and femurs) were examined at 6 mo and 1 year. Spleens were also examined at 1 year. The 25 ppm groups (and corresponding controls) were examined similarly, except that no animals were killed at 6 mo. An abrupt rise in tissue chromium concentrations was noted in rats treated with greater than 5 ppm. The authors stated that “apparently, tissues can accumulate considerable quantities of chromium before pathological changes result.” In the 25 ppm treatment groups, tissue concentrations of chromium were approximately nine times higher for those treated with hexavalent chromium than for the animals exposed to trivalent chromium.

Table 3. Subchronic oral toxicity of hexavalent chromium in rats

Number of animals	Dose and compound	Period of exposure	Endpoints monitored and effect
9 females, 12 males at 25 ppm 10 males, 19 females at 9 ppm 8 males, 8 females at other treatment levels	0, 0.45, 2.2, 4.5, 7.7, 11, 25 ppm as potassium dichromate in drinking water	1 year	No effect based on body weight, gross external condition, histopathological analysis, and blood chemistry.

Source: MacKenzie et al., 1958.

Gross and Heller (1946) reported that 0.125% K_2CrO_4 in the feed of rats was tolerated without observable effects. A dose of 0.25% in the diet resulted in "subnormal condition," including rough coat and "subnormal" young born to treated animals. Doses of 0.5% and 1% in feed resulted in diarrhea, rough dirty coats, and sterility. $ZnCrO_4$ administered in the feed at levels of 0.125%, 0.25%, 0.5%, and 1.0% resulted in subnormal appearance, rough and dirty coats, and sterility at all dose levels. Group sizes, duration of treatment, and criteria for determining sterility were not reported.

4.2.3. Chronic Inhalation Studies

Two studies have provided suggestive evidence of carcinogenicity in mice and rats following inhalation of hexavalent chromium (Nettesheim et al., 1971; Glaser et al., 1986). Nettesheim et al. (1971) exposed C57B1 mice to 13 mg calcium chromate/m^3 (4.33 mg Cr[VI]/m^3 as calcium chromate dust), 5 hours/day, 5 days/week for life. Chromium exposure resulted in a cessation of body weight gain at 6 mo followed by a decrease in body weight thereafter. Bronchial epithelial effects ranged from marked hyperplasia and atrophy to necrosis, suggesting that the maximum tolerated dose may have been exceeded in this study. Inflammatory infiltration into the subepithelium including proliferation of bronchial epithelial cells was noted. Distension of terminal bronchioli and alveoli resembling emphysema was associated with alveolar proteinosis. A 2.8% increase in the number of lung tumors was reported with respect to controls. However, statistical analysis was not performed, and the significance of these results is unclear. In a review of this study, IARC (1990) concluded that a significant excess of treatment-related tumors was not observed. Nettesheim identified 4.33 mg of calcium chromate dust/m^3 as a LOAEL for the occurrence of epithelial necrosis, marked hyperplasia and atrophy of the pulmonary bronchi, emphysema-like changes, and atrophy of the spleen and liver. The single dose used in this study raises the question whether 4.33 mg Cr(VI)/m^3 actually represents a LOAEL.

Glaser et al. (1986) exposed male Wistar rats (20/group) to aerosols of sodium chromate at measured concentrations of 0.025, 0.05, and 0.1 mg Cr(VI)/m^3, 22 hr/day, 7 days/week for 18 mo. An additional group was exposed to a pyrolized Cr(VI)/Cr(III) (3:2) oxide mixture at 0.063 Cr(VI)/m^3. The animals were held under conventional conditions for 12 mo following the exposure. Body weights and mortality were similar among all exposure groups and controls. Primary lung tumors (1 adenocarcinoma and 2 adenomas) and 1 squamous cell carcinoma of the pharynx were evident in the high-chromate exposure group. It is not clear whether the adenocarcinoma and adenomas occurred in the same animal or in different animals. One primary lung adenoma was observed in the oxide-exposed group. Primary lung tumors were not observed in the controls or low-chromate exposure groups. Dose-dependent retention of chromium was seen in the chromate- and oxide-exposed groups relative to the controls. At the conclusion of the study, lung chromium retention was 10-fold greater in the oxide-exposed group than in the high-chromate-exposed group. Significantly increased lung weights were determined for the oxide-exposed group and greater liver weights in the high-exposure group of the chromate. Pigment-loaded macrophages were found in a dose-dependent manner in rats exposed to chromate and to the oxide. Oxide-exposed rats developed focal thickened septa, partially combined with interstitial fibrosis and accumulation of eosinophilic substance in the alveolar lumens. Oxide-exposed groups also demonstrated elevated white- and red-blood cell counts, elevated serum cholesterol, and decreased total serum immunoglobulin levels. The results of this study may provide evidence of a weak carcinogenic potency of aerosols of both sodium dichromate and Cr(VI/III) oxide (Glaser et al., 1986; Glaser et al., 1988).

Steinhoff et al. (1983) investigated the carcinogenicity of soluble sodium dichromate and calcium chromate in Sprague-Dawley rats via intratracheal administration and reported positive carcinogenic effects. The study consisted of 10 treatment groups, one negative control group, and two positive control groups. Each test group contained 40 male and 40 female rats (10 weeks old at the outset). The design of the dose levels selected was such as to assess the impacts of the chemicals delivered in single high doses or in the same dose distributed over a 5-day period. The duration of the study was 2 years and 8 months. Doses ranged from 0.5 to 1.25 mg/kg. Rats administered sodium dichromate or calcium chromate one or five times per week had no significant reduction in survival periods as compared to controls, except in the case of females treated with calcium chromate 5 × 0.25 mg/kg/week. An increased incidence of lung tumors as compared to controls was observed in the treated group in which sodium dichromate was administered in a single dose of 1.25 mg. No lung tumors were observed in the other sodium dichromate treatment groups. In rats administered calcium chromate, statistically significant increases in lung tumors were found in groups treated with a single dose of 1.25 mg/kg as well as in the group treated with 5 × 0.25 mg/kg/week distributed over a period of 5 days.

There is some evidence that hexavalent chromium may be carcinogenic following intrapleural implantation of calcium chromate (Hueper and Payne, 1962) or intrabronchial implantation of strontium chromate, calcium chromate, or zinc chromate (Levy and Martin, 1983). These tumors, however, were observed only at the site of implantation. Steffee and Baetjer (1965) observed statistically significant increases in lung tumors following intratracheal

instillation of 0.01 to 0.03 mg zinc chromate in strain A mice. The instillations were performed at 2-week intervals and the animals were observed until death.

In contrast, intratracheal and intrapleural implantation studies of other chromium compounds have not demonstrated increases in tumor incidences. Mixed hexavalent and trivalent chromium-containing dust was not carcinogenic in strain A Swiss and C57BL mice and mixed-breed rats following intratracheal implantation (Baetjer et al., 1959). Steffee and Baetjer (1965) did not observe increases in lung adenomas following instillation of chromium dust, zinc chromate, and lead chromate into the tracheas of guinea pigs and rabbits. Hueper and Payne (1962) reported similar negative results after instillation of strontium chromate or calcium chromate suspended in gelatin; however, the experimental detail in the report was insufficient for adequate evaluation. Hueper and Payne (Hueper, 1955, 1958; Payne, 1960; Hueper and Payne, 1962) described a series of studies in rats treated by intrapleural injection of a number of hexavalent or trivalent compounds. Hueper (1955) injected powdered metallic chromium into the pleural cavity of rats, guinea pigs, and mice and observed no significant increase in tumor incidence, either at the injection site or in other organs. Payne (1960) implanted chromite roast, from which the soluble sodium chromate was extracted, into the pleural cavity of 35 rats. None of the 35 control animals developed tumors, and three of the treated animals developed tumors at the implantation site. In an earlier study, Hueper (1958) using chromite roast not leached of sodium chromate, none of the 25 treated male Bethesda rats developed implantation site tumors during 24 mo; however, the early deaths of nine of the treated animals decreased the number of animals at risk. Hueper and Payne (1962) noted that no implantation site tumors were observed in 42 rats during a 24-mo period following eight implantations of 25 mg of trivalent chromium acetate in gelatin over a 13-mo period.

Baetjer et al. (1959) chronically exposed three strains of mice (Strain A, Swiss, and C57Bl) and mixed-breed rats to approximately l mg chromium dust/m^3, 4 hours/day, 5 days/week over a range of 16-58 weeks, and reported no increase in the incidence of lung tumors with respect to untreated controls. Similar results were obtained by Steffee and Baetjer (1965) for Wistar rats, rabbits, and guinea pigs exposed to chromium dust.

Laskin (1972) exposed rats and hamsters to calcium chromate aerosol at a level of 2 mg/m^3 (0.67 mg Cr(VI)/m^3) for 589 days out of an 891-day study. Although some laryngeal hyperplasias and metaplasias were observed in both species tested, details pertaining to controls were not given in the available review.

4.2.4. Subchronic Inhalation Studies

Data from animal studies identify the respiratory tract as the primary target of chromium toxicity following inhalation. Glaser et al. (1985) exposed 5-week-old male Wistar rats to aerosols of sodium dichromate at concentrations ranging from 0.025 to 0.2 mg Cr(VI)/m^3, 22 hr/day in subacute (28 day) or subchronic (90 day) protocols. Subacute and subchronic exposures to Cr(VI) aerosol concentrations resulted in a positive correlation between exposure dose and significant effects on alveolar macrophages and immunological function. Inhalation of Cr(VI) aerosols stimulated the humoral immune system. Differences in the mean total serum

immunoglobulin were significant at exposures above 0.025 mg/m^3, while exposures to aerosol concentrations greater than 0.1 mg/m^3 resulted in depression of the immune system stimulation. The primary antibody response to the β-cell-dependent antigen sheep red blood cell was elevated in a chromium time- and dose-dependent manner. The immune stimulating effect of subchronic exposure to an aerosol with 0.05 mg/m^3 chromium was not reversed after 2 months of fresh air regeneration. Subchronic exposure to 0.2 mg/m^3 chromium resulted in depression of the immune-stimulating effect relative to the response at 0.05 mg/m^3. The spleen T-lymphocyte subpopulation was also stimulated by subchronic exposure to 0.2 mg/m^3 chromium. Bronchoalveolar lavage (BAL) cell counts were significantly decreased following subchronic exposure to levels above 0.025 mg/m^3 chromium, though it was not clear whether the effectiveness of the lavagability of the cells was altered at the higher dose levels. The number of lymphocytes and granulocytes showed a slight but significant increase in the lavage fluids of the of the subacute and subchronically exposed groups. At subacute exposure concentrations up to 0.05 mg/m^3 the phagocytic activity of the alveolar macrophages increased; however, subchronic exposure at 0.2 mg/m^3 decreased this function significantly, complicating the interpretation of this result. Following subacute exposure to 0.2 mg/m^3 chromium, reductions in macrophage cell counts and phagocytic activities correlate with an observed lower clearance of inhaled iron oxide. Inhaled chromium was found to preferentially accumulate in the lung following exposure to chromate aerosols. Lung and spleen weights were significantly increased ($p < 0.005$) after both subacute and subchronic inhalation of chromate aerosols at concentrations greater than 0.025 mg/m^3. Serum contents of triglycerides and phospholipids differed significantly from controls ($p < 0.05$) in rats exposed subchronically to 0.2 mg/m^3 chromate.

Glaser et al. (1990) presented a paper at the Second European Meeting of Environmental Hygiene that reported exposure of 8-week-old male Wistar rats to sodium dichromate at 0.05, 0.1, 0.2, and 0.4 mg Cr(VI)/m^3 22 hr/day, 7 days/week for 30-90 days. Chromium-induced effects occurred in a strong dose-dependent manner. The authors observed obstructive respiratory dyspnea and reduced body weight following subacute exposure at the higher dose levels. The mean white blood cell count was increased at all doses ($p < 0.05$) and was related to significant dose-dependent leukocytosis following subacute exposures. Mean lung weights were significantly increased at exposure levels of 0.1 mg/m^3 following both the subacute and subchronic exposures. Accumulation of macrophages was seen in all of the exposure groups and was postulated to be a chromium-specific irritation effect that accounted for the observed increases in lung weights.

Focal inflammation was observed in the upper airways following the subchronic exposure. BAL analyses provided more detailed information on the nature of the dichromate-induced irritation effect. BAL albumin was increased following the subacute exposure, and was taken to indicate exudation into the alveolar region as an early irritation effect. The mean protein content of the cell-free lavage fluid was significantly increased in a dose-dependent fashion after the subacute and subchronic exposures. However, protein levels returned to control levels following a recovery period. Cytosolic lactate dehydrogenase and the number of mononuclear macrophages were also elevated following the subacute and subchronic exposures, particularly at the highest dose levels. The enzyme activity and number of macrophages returned to the control level following the recovery period. The authors concluded that chromium inhalation induced

pneumocyte toxicity and suggested that inflammation is essential for the induction of most chromium inhalation effects and may influence the carcinogenicity of Cr(VI) compounds (Glaser et al., 1990).

Johansson et al. (1980) exposed groups of four rabbits to chromium dust at concentrations of 3.1 mg/m^3 and 0.6 mg/m^3 for 5 days/week, 6 hours/day for 4 weeks. Macrophages collected from rabbits exposed to the higher concentration of chromium phagocytized significantly more chromium particles than the controls, though the number of nonviable macrophages was less than 3%.

Johansson et al. (1986) exposed groups of rabbits to aerosols of hexavalent (0.9 mg Cr[VI]/m^3 as Na_2CrO_4) or trivalent (0.6 mg Cr[III]/m^3 as $Cr[NO_3]_3$) chromium 5 days/week, 6 hours/day for four to six weeks. The number of macrophages obtained from the lungs of the rabbits exposed to Cr(VI) was significantly increased. While the numbers of macrophages from rabbits exposed to Cr(III) were not increased, striking morphological changes were observed, including round dark chromium-rich inclusions in the cytoplasm, an increased number of cells with a smooth, inactive cell surface, enlarged Golgi apparatus, and a tendency toward elongated cell shape. The macrophages from rabbits exposed to Cr(VI) showed less marked morphological changes than those exposed to Cr(III).

Lee et al. (1988) exposed groups of 30 male and 30 female rats to 0.5 mg/m^3 or 25 mg/m^3 CrO_2 (IV) 6 hr/day, 5 days/week for 2 years. There were no compound-related differences in weight gain between exposed and control groups and no exposure-related mortality in any exposed group. There were no compound-related lesions in the vital organs and tissues other than in the lungs of exposed rats. Dust-laden alveolar macrophages with slight Type II pneumocyte hyperplasia were noted following exposure at 0.5 mg/m^3. Inhaled particles were deposited mainly in the alveoli adjacent to the alveolar ducts and the dust particles appeared as dense particles and were phagocytized by intraalveolar macrophages. Exposure at 25 mg/m^3 was suggested to have overwhelmed the lung clearance mechanisms and resulted in significant increases in dust-laden macrophages, bronchoalveolar cell hyperplasia with foamy macrophage response, and cholesterol granuloma. The mechanism of action was not confirmed in the study, and the observation of dust particles in the alveolar macrophages is not necessarily indicative of overloading of the lung clearance mechanism. At 24 mo, the dust deposition and effects were increased significantly, with severe dust-laden macrophages, dust deposition in peribronchial lymphoid tissue, hyperplasia of Type II pneumocytes, and collagenized fibrosis occurring in 100% of all lung tissues examined in either gender. Two female rats developed well-differentiated cystic keratinizing squamous cell carcinomas with no tumor metastasis. The tumors were not characterized as neoplastic lesions.

Mice exposed to 1.81 or 3.63 mg/m^3 Cr(VI) as CrO_3 for 1 year developed nasal septal perforation, loss of cilia, and metaplasia of the lung, trachea, and bronchus (Adachi, 1987; Adachi et al., 1986)

4.3. REPRODUCTIVE/DEVELOPMENTAL STUDIES

4.3.1. Oral Studies

High doses of Cr(VI) compounds have been reported to cause developmental toxicity in mice. Trivedi et al. (1989) exposed mice to 250, 500, and 1,000 ppm potassium dichromate daily through drinking water during the entire gestational period. The authors reported decreased fetal weight, increased resorptions, and increased abnormalities (tail kinking, delayed ossification of the cranium) in exposed mice. The medium- and high-dose groups registered significant reductions in body weight gain when compared to controls. The most significant finding of the study was the complete absence of uterine implantation in the high-dose group. The 250 and 500 ppm dose groups also showed significant incidences of resorption as compared to controls. The authors observed significant increases in preimplantation and postimplantation losses and dose-dependent reductions in total weight and crown-rump length in the lower dose groups. Additional effects included treatment-related increases in abnormalities in the tail, wrist forelimbs and subdermal hemorrhagic patches in the offspring.

Zahid et al. (1990) fed BALB/C albino Swiss mice trivalent (chromium disulfate) and hexavalent (potassium dichromate) chromium at concentrations of 100, 200, and 400 ppm for 35 days in the diet. The authors stated that the exposure groups included seven animals per group, and an additional seven animals were used as controls, though conflicting summaries of the actual group sizes are presented throughout the report. Following the treatment, the authors examined the testes and epididymis of the animals. The epididymis was weighed and minced suspended in buffered formalin. Sperm counts were then subsequently determined and sperms were examined for morphological abnormalities. Testes were fixed with Bouin's fluid for 1 week and were subsequently sectioned to 0.6 micron thickness and stained with haematoxylin and eosin for histological examination. Ten sections were chosen randomly from the anterior, middle, and posterior parts of each testis and studied. One seminiferous tubule was chosen and examined to determine the cellular stages of spermatogenesis and the number of degenerated tubules. Statistical analyses of the data were conducted using the *t*-test between means and the 2×2 contingency chi-square test between percentages. The authors reported deleterious effects on the male mouse testes, including ambiguous levels of degeneration in the outermost cellular layers of the seminiferous tubules, reduced (or absent) spermatogonia per tubule, accumulation of germ cells in the resting spermatocytes stage, reduced sperm count in the epididymis, and increased percentage of morphologically abnormal sperms at all dose levels. The authors concluded that the small but significant increase of hexavalent chromium in the testes of fed animals induced significant degeneration.

Serious questions have been raised regarding the design and conduct of this study (Finley et al., 1993; NTP 1996a,b, 1997). The methods utilized in the Zahid et al. study are considered to be insufficient to identify spermatogonia, likely generated nonreproducible counts of epididymal sperm, and resulted in the biologically implausible conclusion of reduction in spermatogonia numbers concurrent with unchanged spermatocyte and spermatid numbers. Additional questions have been raised with regard to uncertainties regarding the actual groupings of animals used and the statistical analysis of the data.

The National Toxicology Program recently conducted a three-part study to investigate oral ingestion of hexavalent chromium in experimental animals (NTP, 1996a,b, 1997). The study included a determination of the potential reproductive toxicity of potassium dichromate in Sprague-Dawley rats, a repeat of the study of Zahid et al. (1990) using BALB/C mice, and a Reproductive Assessment by Continuous Breeding study in BALB/C mice.

The study in the Sprague-Dawley rat (NTP, 1996a) was conducted in order to generate data in a species commonly used for regulatory studies. Groups of 24 males and 48 females were exposed to 0, 15, 50, 100, or 400 ppm potassium dichromate daily in the diet for 9 weeks followed by a recovery period of 8 weeks. Six male and 12 female rats were sacrificed after 3, 6, or 9 full weeks of treatment or after the full recovery period. Animals were examined for body weights; feed and water consumption; organ weights; microscopic evaluation of the liver, kidney, and ovaries; hematology; histology of the testis and epididymus for Sertoli nuclei and preleptotene spermatocyte counts in Stage X or XI tubules; and chromatin analysis. No treatment-related hematology findings were reported except for slight decreases in MCV and MCH values in the male and female treatment groups receiving 400 ppm potassium dichromate (24 mg/kg-day). While the trends in MCV and MCH were not large and were within the reference ranges (Charles River Laboratories, 1993), they are consistent with the findings of the companion studies in BALB/C mice (see below) and were characterized by the authors as suggestive of a potential bone marrow/erythroid response. The authors considered the 100 ppm (6 mg/kg-day) dose group to be representative of the NOAEL for the study.

The reproductive study in BALB/C mice (NTP, 1996b) was conducted to reproduce the conditions utilized by Zahid et al. (1990) in their examination of comparative effects of trivalent and hexavalent chromium on spermatogenesis of the mouse. Groups of 24 male and 48 female BALB/C mice were exposed to 0, 15, 50, 100, or 400 ppm potassium dichromate in the diet for 9 weeks followed by a recovery period of 8 weeks. Six male and 12 female mice were sacrificed after 3, 6, or 9 full weeks of treatment or after the full recovery period. Animals were examined for body weights; feed and water consumption; organ weights; microscopic evaluation of the liver, kidney, and ovaries; hematology; histology of the testis and epididymus for Sertoli nuclei and preleptotene spermatocyte counts in Stage X or XI tubules; and chromatin analysis. Treatment-related effects included a slight reduction in the mean body weights in the 400 ppm males and the 100 ppm females, a slight increase in food consumption at all dose levels, a slight decrease in MCV and MCH at 400 ppm, and cytoplasmic vacuolization of the hepatocyte at 50, 100 and 400 ppm. None of the effects on spermatogenesis reported by Zahid et al. (1990) were observed in this study. On the basis of the cytoplasmic vacuolization of the hepatocyte in the 50, 100, and 400 ppm dose groups, the authors selected 15 ppm (4 mg/kg-day) as the NOAEL.

The potential reproductive toxicity of potassium dichromate was further evaluated in BALB/C mice using the Reproductive Assessment by Continuous Breeding protocol (NTP, 1997). In the continuous breeding phase of the study, groups of 20 male and female pairs of animals (F_0) were exposed to dose levels of 0, 100, 200, and 400 ppm potassium dichromate, based on the previous study of reproductive effects in BALB/C mice. Litters born after the continuous breeding phase (F_1) received the same concentrations of potassium dichromate as their F_0 parents. F_1 animals were used for assessment of second-generation reproductive toxicity.

At sexual maturity, 20 control animals of each sex and 20 treated animals of each sex in each dose group were selected as breeding pairs (avoiding sibling matings), cohabitated for 7 days, and then separated. Offspring were counted and examined for terminal body and organ weights and received sperm and tissue analysis. The NTP studies were designed to repeat the findings of Zahid using methods of greater rigor and definition, but were unable to do so. The reproductive assessment indicated that potassium dichromate administered at 100, 200, or 400 ppm in the diet to male and female BALB/C mice is not a reproductive toxicant in either sex. Fertility and the pregnancy index (number delivering/number cohabitated) were not affected by continued exposure to potassium dichromate. No treatment-related differences were observed in the mean average litters per pair, number of live pups per litter, proportion of pups born alive, sex ratio, absolute live pup weight, or adjusted live pup weight. Mean body weights of the high-dose F_0 and F_1 animals were slightly decreased relative to controls, and mean food consumption in the F_1 animals was increased relative to controls. Mean absolute liver weights in the high-dose group F_0 animals were decreased relative to controls. Treatment-related changes were observed in the hematology data for the F_1 animals. The MCV was slightly decreased in the high-dose males, and the MCH was slightly decreased in the female animals in all dose groups. The authors suggested that the NOAEL was not established in this study because of the slight decrease in MCH in the females of the 100 ppm (22.4 mg/kg-day) dose group.

Junaid et al. (1996) exposed female Swiss albino mice to 250, 500, or 750 ppm potassium dichromate in drinking water to determine the potential embryotoxicity of hexavalent chromium during days 6-14 of gestation. No notable changes in behavior or clinical signs were observed in the control or treated dams. Chromium levels in blood, placenta, and fetus increased in a dose-dependent fashion over the course of the study. The authors reported retarded fetal development and embryo- and fetotoxic effects including reduced fetal weight, reduced number of fetuses (live and dead) per dam, and higher incidences of stillbirths and postimplantation loss in the 500 and 750 ppm dosed mothers. Significantly reduced ossification in nasal, frontal, parietal, interparietal, caudal, and tarsal bones was observed in the high-dose group, while reduced ossification in only the caudal bones was observed in the 500 ppm dose group. Based on the body weight of the animals (30 +/- 5 g) and the drinking water ingested by the animals in the 250 ppm dose group (8.0 ml/mouse/day), the dose level in the 250 ppm group can be identified as 67 mg/kg-day.

Kanojia et al. (1996) exposed female Swiss albino rats to 250, 500, or 750 ppm potassium dichromate in drinking water to determine the potential teratogenicity of hexavalent chromium pregestationally for 20 days. No notable changes in behavior or clinical signs were observed in the control or treated dams. Chromium levels in blood, placenta, and fetus were significantly increased in the dams of the 500 and 750 ppm dose groups. The authors reported a reduced number of corpora lutea and implantations, retarded fetal development, and embryo- and fetotoxic effects including reduced number of fetuses (live and dead) per dam and higher incidences of stillbirths and postimplantation loss in the 500 and 750 ppm dosed mothers. Significantly reduced parietal and interparietal ossification was observed in the high-dose group. Based on the body weight of the animals (175 +/- 25 g) and the drinking water ingested by the animals in the 250 ppm dose group (26 ml/mouse/day) the dose level in the 250 ppm group can be identified as 37 mg/kg-day.

Elbetieha and Al-Hamood (1997) examined fertility following potassium dichromate exposures in mice at concentrations considerably greater than those used by NTP. Sexually mature male and female mice were exposed to 1,000, 2,000, 4,000, or 5,000 mg/L potassium dichromate in drinking water for 12 weeks. The effects of the exposures on fertility was examined at 140 days. No mortality or clinical signs of toxicity were reported in any group of male or female mice exposed at any concentration in the experiment. Exposure of male mice to hexavalent chromium compounds for 12 weeks had adverse impacts on the male reproductive system and fertility, though the mating capability of the mice was not affected. Fertility was significantly reduced in males exposed to 5,000 mg/L potassium dichromate. Testes weights were significantly increased in the males exposed in the 2,000 and 5,000 mg/L dose groups, while seminal vesicle and preputial gland weights were significantly reduced in the 5,000 mg/L exposed males. The numbers of implantation sites and viable fetuses were significantly reduced in females impregnated by males exposed to 2,000 and 4,000 mg/L. The numbers of implantations and viable fetuses were significantly reduced in pregnant females exposed to 2,000, 4,000, and 5,000 mg/L of the hexavalent chromium compound.

The findings of Junaid et al. (1996) and Kanojia et al. (1996) are consistent with those of Trivedi et al. (1989), and studies suggest the presence of embryotoxic and fetotoxic effects of potassium dichromate following oral exposures in mice and rats. The studies utilized similar dose levels provided in the drinking water of female mice and rats. The dose levels are similar to those used by the NTP study, which demonstrated no reproductive effects following administration of potassium dichromate in the diet. It cannot be determined whether the lack of reproductive toxicity demonstrated in the NTP studies are reflective of the reduced bioavailability of hexavalent chromium provided in the diet in comparison to that provided in drinking water, or whether the NTP studies identified a NOAEL for reproductive effects.

4.3.2. Inhalation Studies

No developmental effects were reported in rats exposed to 0.2 mg/m^3 Cr(VI) as sodium dichromate for three generations (Glaser et al., 1984). No histopathological effects of the testes were reported following exposure of rats to 0.2 mg/m^3 Cr(VI) as sodium dichromate for 28 or 90 days (Glaser et al., 1985) or 0.1 mg/m^3 Cr(VI) as sodium dichromate for 18 mo (Glaser et al., 1986, 1988).

4.4. OTHER STUDIES

4.4.1. Contact Dermatitis

Chromium is one of the most common contact sensitizers in males in industrialized countries (Fowler, 1990; Cronin, 1980) and is associated with occupational exposures to numerous materials and processes, including chromeplating baths, chrome colors and dyes, cement, tanning agents, wood preservatives, anticorrosive agents, welding fumes, lubricating oils and greases, cleaning materials, and textiles and furs (Burrows and Adams, 1990; Polak et al., 1973). Solubility and pH appear to be the primary determinants of the capacity of individual

chromium compounds to elicit an allergic response (Fregert, 1981; Polak et al., 1973). The low solubility Cr(III) compounds are much less efficient contact allergens than Cr(VI) (Spruit and van Neer, 1966).

Dermal exposure to chromium has been demonstrated to produce irritant and allergic contact dermatitis (Bruynzeel et al., 1988; Polak, 1983; Cronin, 1980; Hunter, 1974). Primary irritant dermatitis is related to the direct cytotoxic properties of chromium, while allergic contact dermatitis is an inflammatory response mediated by the immune system. Allergic contact dermatitis is a cell-mediated immune response that occurs in a two-step process. In the first step (induction), chromium is absorbed into the skin and triggers an immune response (sensitization). Sensitized individuals will exhibit an allergic dermatitis response when exposed to chromium above a threshold level (Polak, 1983). Induction is generally considered to be irreversible. Chromium allergic dermatitis is characterized by symptoms of erythema, swelling, papules, small vesicles, dryness, scaling, and fissuring (Adams, 1990; MacKie, 1981).

4.4.2. Toxicant Interactions

Potassium dichromate has been reported to potentiate the effects of the nephrotoxins, mercuric chloride, citrinin, hexachlorobutadiene, and maleic acid (ATSDR, 1993). The genotoxicity of hexavalent chromium has also been shown to be altered in the presence of other compounds, including ascorbic acid and vitamin E and thiol compounds (Susa et al., 1994). Vitamin B_2 has been reported to enhance the cytotoxicity of sodium chromate (ATSDR, 1993).

4.4.3. Genotoxicity

Hexavalent chromium is rapidly taken up by cells through the sulfate transport system (Sugiyama, 1992). Once inside the cell, Cr(VI) is quickly reduced to the trivalent form by cellular reductants, including ascorbic acid, glutathione, and flavoenzymes, such as cytochrome P-450 glutathione reductase, and riboflavin. The intracellular reduction of Cr(VI) generates reactive chromium V and chromium IV intermediates as well as hydroxyl free radicals ($^{\bullet}OH$) and singlet oxygen ($^1O_2$). A variety of DNA lesions are generated during the reduction of Cr(VI) to Cr(III), including DNA strand breaks, alkali-labile sites, DNA-protein and DNA-DNA crosslinks, and oxidative DNA damage, such as 8-oxo-deoxyguanosine. The relative importance of the different chromium complexes and oxidative DNA damage in the toxicity of Cr(VI) is unknown.

Hexavalent chromium has been shown to be genotoxic only in the presence of appropriate reducing agents in vitro or in viable cell systems in vitro or in vivo. Hexavalent chromium has been shown to be mutagenic in bacterial systems in the absence of a mammalian activating system (Venitt and Levy, 1974; Nishioka, 1975; Nakamuro et al., 1978; Green et al., 1976; Kanematsu et al., 1980; Lofroth and Ames, 1978; Newbold et al., 1979; Bonatti et al., 1976; Fukanaga et al., 1982), and not mutagenic when a mammalian activating system is present (Lofroth, 1978; Petrilli and DeFlora, 1977, 1978a,b). Hexavalent chromium is also mutagenic in eukaryotic test systems (Bonatti et al., 1976; Newbold et al., 1979; Fukanaga et al., 1982) and clastogenic in cultured mammalian cells (Raffetto, 1977; Levis and Majone, 1979; Umeda and

Nishimura, 1979; Tsuda and Kato, 1977; Newbold et al., 1979; Nakamuro et al., 1978; Stella et al., 1982; Ohno et al., 1982; Gomez-Arroyo et al., 1981; Wild, 1978; Sarto et al., 1982). Hexavalent chromium in the presence of glutathione has been demonstrated to produce genotoxic DNA adducts that inhibit DNA replication and are mutagenic (Snow, 1994).

4.5. SYNTHESIS AND EVALUATION OF MAJOR NONCANCER EFFECTS

4.5.1. Oral Studies

4.5.1.1. *Human Studies*

Cr(VI) is considerably more toxic than Cr(III). A cross-sectional study reported the effects of environmental contamination of well water adjacent to a chromium alloy plant. Cr(VI) concentrations were reported as 20 mg/L, with an estimated exposure dose of 0.57 mg/kg-day (Zhang and Li, 1987) No lower dose levels were reported in this study. Reported effects included oral ulcers, diarrhea, abdominal pain, indigestion, vomiting, leukocytosis, and presence of immature neutrophils. The single high dose level reported in the study limits its usefulness for quantitative risk assessment purposes.

Other reports of toxic effects in humans are limited to case reports from accidental poisonings. Some Cr(VI) compounds are potent oxidizing agents (such as potassium tetrachromate and chromium trioxide) and are thus strong irritants of mucosal tissue. Effects included metabolic acidosis, acute tubular necrosis, kidney failure, and death (Saryan and Reedy, 1988).

4.5.1.2. *Animal Studies*

Only one chronic study pertaining to the oral toxicity of hexavalent chromium was located in the available literature. Anwar et al. (1961) exposed dogs orally (2 dogs/group) to potassium chromate in drinking water for 4 years. No effects were observed with regard to gross and microscopic analysis of all major organs, urinalysis, and weights of spleen, liver, and kidney. A NOEL of 0.31 mg potassium chromate/kg/day can be established from this study. The small group size in this study limits its usefulness for quantitative risk assessment purposes.

Mackenzie et al. (1958) exposed groups of male and female rats to potassium dichromate (0-25 ppm of hexavalent chromium) in drinking water for 1 year. No effects were observed at any level of treatment, and a NOEL of 2.5 mg/kg-day can be established based on body weight, gross external condition, histopathological analysis, and blood chemistry.

Junaid et al. (1996) exposed female Swiss albino mice to 250, 500, or 750 ppm potassium dichromate in drinking water to determine the potential embryotoxicity of hexavalent chromium during days 6-14 of gestation. The authors reported retarded fetal development and embryo- and fetotoxic effects including reduced fetal weight, reduced number of fetuses (live and dead) per dam, and higher incidences of stillbirths and postimplantation loss in the 500 and 750 ppm dosed

mothers. Significantly reduced ossification in bones was also observed in the medium- and high-dose groups. Based on the body weight of the animals (30 +/- 5 g) and the drinking water ingested by the animals in the 250 ppm dose group (8.0 ml/mouse/day), the dose level in the 250 ppm group can be identified as 67 mg/kg-day.

Kanojia et al. (1996) exposed female Swiss albino rats to 250, 500, or 750 ppm potassium dichromate in drinking water to determine the potential teratogenicity of hexavalent chromium pregestationally for 20 days. The authors reported a reduced number of corpora lutea and implantations, retarded fetal development, and embryo- and fetotoxic effects including reduced number of fetuses (live and dead) per dam and higher incidences of stillbirths and postimplantation loss in the 500 and 750 ppm dosed mothers. Significantly reduced parietal and interparietal ossification was observed in the high-dose group. Based on the body weight of the animals (175 +/- 25 g) and the drinking water ingested by the animals in the 250 ppm dose group (26 ml/mouse/day), the dose level in the 250 ppm group can be identified as 37 mg/kg-day.

Elbetieha and Al-Hamood (1997) exposed sexually mature male and female mice to 1,000, 2,000, 4,000, or 5,000 mg/L potassium dichromate in drinking water for 12 weeks. The effects of the exposures on fertility was examined at 140 days. The authors reported adverse impacts on the male reproductive system and fertility, though the mating capability of the mice was not affected. Testes weights were significantly increased in the males exposed in the 2,000 and 5,000 mg/L dose groups, while seminal vesicle and preputial gland weights were significantly reduced in the 5,000 mg/L exposed males. The number of implantation sites and viable fetuses was significantly reduced in females impregnated by males exposed to 2,000 and 4,000 mg/L, and the numbers of implantations and viable fetuses was significantly reduced in pregnant females exposed to 2,000, 4,000, and 5,000 mg/L of the hexavalent chromium compound. Information regarding the amount of water consumed by the animals was not provided in this study.

4.5.2. Inhalation Studies

4.5.2.1. *Human Studies*

4.5.2.1.1. ***Respiratory tract effects*****.** Three studies on chromeplaters provide some quantitative information on upper respiratory irritation after exposure to Cr(VI) as chromic acid. In the study of Cohen et al. (1974), nasal ulcers and perforations were associated with total chromium concentrations of 1.4 to 43.9 µg/m^3, averaging 7.1 µg/m^3, and Cr(VI) concentrations of 0.09 to 9.1 µg/m^3, averaging 2.9 µg/m^3. Ninety-five percent of the 37 workers studied exhibited pathologic changes in nasal mucosa in a concentration-duration response. More than half of the workers employed less than 1 year had nasal pathology that was more severe than simple redness of the nasal mucosa. Almost all the workers (35 of 37) employed longer than 1 year had nasal tissue damage. The authors noted the lack of good industrial hygiene practices, implicating direct contact, such as touching of the nose with chromium-contaminated hands, as a potentially important route of exposure. A subsequent study by Lucas and Kramkowski (1975) revealed similar results. Cr(VI) concentrations ranged from 1 to 20 µg/m^3, averaging 4 µg/m^3. The authors attributed the nasal pathology primarily to direct contact. Lindberg and Hedenstierna

(1983) also found similar effects on nasal pathology and subjective symptoms. They reported reddening of the nasal mucosa at 1 to 2 $\mu g/m^3$, and nasal irritation (chronic and nasal septal ulceration and perforation) in two-thirds of the subjects exposed to concentrations of 2 to 20 $\mu g/m^3$. All workers with nasal ulceration had been exposed to chrome acid mist, which contained Cr(VI) at 20 $\mu g/m^3$, or greater than 20 $\mu g/m^3$ near the baths. Changes in pulmonary function (vital capacity and forced expiatory volume) were seen at Cr(VI) exposures greater than 2 $\mu g/m^3$.

4.5.2.1.2. ***Renal effects.*** Exposure to Cr(VI) at concentrations as low as 4 to 6 $\mu g/m^3$ has been reported to result in elevated excretion of ß-2-microglobulin (Lindberg and Vesterberg, 1983b). The effect may be reversible because former chromeplaters did not have an elevated concentration of either ß-2-microglobulin or albumin in their urine. Saner et al. (1984) did not find increased urinary ß-2-microglobulin levels in tannery workers in comparison to referent control workers. However, urinary chromium concentrations in the Saner et al. study were likely distinctly lower than those in the study of Lindberg and Vesterberg.

In summary, effects on the airways and kidney have been observed in chromeplaters exposed subchronically to chromic acid mist containing Cr(VI) in air at concentrations greater than 1 $\mu g/m^3$. Such effects include reddening of nasal mucosa, nasal irritation (ulceration, perforation), changes in pulmonary function, and renal proteinuria. Many of the available studies lack quantitative concentration-response data on chromium health effects suitable for quantitative risk assessment.

4.5.2.2. ***Animal Studies***

Data from studies in rats, mice, and rabbits identify the respiratory tract as the primary target of chromium toxicity following inhalation. Glaser et al. (1985) exposed rats to 0-0.2 mg Cr(VI)/m^3 22 hr/day, 7 days/week for 90 days. The authors reported increased lung and spleen weight, and increased macrophage activity and percent lymphocytes in BAL fluid. Glaser et al. (1990) exposed rats to 0-0.4 mg Cr(VI)/m^3, 22 hr/day, 7 days/week for 30-90 days and reported hyperplasia, increased lung weight, macrophage infiltration, and LDH in bronchoalveolar lavage fluid (BALF). The authors suggested that inflammation is essential for the induction of most chromium inhalation effects and may influence the carcinogenicity of Cr(VI) compounds.

Rats exposed to 0.5 mg/m^3 CrO_2 (IV) 6 hr/day, 5 days/week for 2 years produced dust-laden alveolar macrophages with slight Type II pneumocyte hyperplasia. Exposure at 25 mg/m^3 overwhelmed the lung clearance mechanisms and resulted in significant increases in dust-laden macrophages, bronchoalveolar cell hyperplasia with foamy macrophage response, and cholesterol granuloma (Lee et al., 1988).

Mice exposed to 1.81 or 3.63 mg/m^3 Cr(VI) as CrO_3 for 1 year developed nasal septal perforation, loss of cilia, and metaplasia of the lung, trachea, and bronchus (Adachi, 1987; Adachi et al., 1986). Epithelial changes of the bronchial tree ranging from necrosis and atrophy to hyperplasia were observed in mice exposed to 4.3 Cr/m^3 as $CaCrO_4$ dust for 18 months (Nettesheim et al., 1971).

Rabbits exposed to chromium dust at 3.1 mg/m^3 and 0.6 mg/m^3 for 5 days/week, 6 hours/day for 4 weeks phagocytized significantly more chromium particles than the controls, though the number of nonviable macrophages was less than 3% (Johansson et al., 1980). In a subsequent study, rabbits were exposed to aerosols of hexavalent (0.9 mg/m^3 Na_2CrO_4) or trivalent (0.6 mg/m^3 $Cr[NO_3]_3$) chromium for 5 days/week, 6 hours/day for 4 to 6 weeks. The number of macrophages obtained from the lungs of the rabbits exposed to Cr(VI) was significantly increased, while striking morphological changes were observed in macrophages of rabbits exposed to Cr(III) (Johansson et al., 1986).

4.6. WEIGHT-OF-EVIDENCE EVALUATION AND CANCER CHARACTERIZATION

Applying the criteria outlined in the Guidelines for Carcinogen Risk Assessment (U.S. EPA, 1986) for evaluating the overall weight of evidence for carcinogenicity to humans, hexavalent chromium is most appropriately designated a Group A - Known Human Carcinogen. Using the proposed Guidelines for Carcinogen Risk Assessment (U.S. EPA, 1996), hexavalent chromium is most appropriately designated a known human carcinogen by the inhalation route of exposure on the following basis. The potential carcinogenicity of chromium by the oral route of exposure cannot be determined at this time.

Results of occupational epidemiologic studies of chromium-exposed workers are consistent across investigators and study populations. Dose-response relationships have been established for chromium exposure and lung cancer. Workers in the chromium industry are exposed to both Cr(III) and Cr(VI) compounds. Because only Cr(VI) has been found to be carcinogenic in animal studies, however, data support only the classification of Cr(VI) as a human carcinogen.

Animal data provide suggestive evidence of the carcinogenicity of hexavalent chromium. Hexavalent chromium compounds have produced the following tumor types in animal assays: lung tumors following inhalation of aerosols of sodium chromate and pyrolized Cr(VI)/Cr(III) oxide mixtures in rats, lung tumors following intratracheal administration of sodium dichromate in rats, intrapleural implant site tumors for various Cr(VI) compounds in rats, intrabronchial implantation site tumors for various Cr(VI) compounds in rats, intramuscular injection site tumors in rats and mice, and subcutaneous injection site sarcomas in rats. Inflammation is considered to be essential for the induction of most chromium inhalation effects and may influence the carcinogenicity of Cr(VI) compounds (Glaser et al., 1985).

In vitro data are suggestive of a potential mode of action for hexavalent chromium carcinogenesis. Cr(VI) readily passes through cell membranes and is rapidly reduced intracellularly to generate reactive Cr(V) and Cr(IV) intermediates and reactive oxygen species. The reactive oxygen species may interact with DNA to form premutagenic lesions. Hexavalent chromium has been shown to be mutagenic in bacterial assays, yeasts, and V79 cells, and Cr(VI) compounds decrease the fidelity of DNA synthesis in vitro and produce unscheduled DNA synthesis as a consequence of DNA damage. Chromate has been shown to transform both primary cells and cell lines.

IARC (1990) concluded that there is sufficient evidence of respiratory carcinogenicity in humans occupationally exposed during chromate production. Animal data were considered supportive of the epidemiological data; however, the relative contributions to carcinogenic risk of metallic chromium, trivalent chromium, hexavalent chromium, or soluble versus insoluble chromium compounds could not be elucidated. IARC (1982) classified chromium and chromium compounds as Group I chemicals.

At present, the carcinogenicity of hexavalent chromium by the oral route of exposure cannot be determined. One study of miners in Ontario suggested that exposure to chromium may have been associated with stomach cancer, but other human and animal studies have not reported similar effects.

4.7. OTHER HAZARD IDENTIFICATION ISSUES

4.7.1. Possible Childhood Susceptibility

A number of factors may differentially affect the response of children to toxicants such as Cr(VI). These factors include diet and physical environment as well as maturation of physiological and biochemical processes. At present, there is too little information to make any statements about how these factors may specifically affect the toxicological responses of Cr(VI) in children, be they cancer or noncancer.

4.7.2. Possible Sex Differences

The extent to which men differ from women in susceptibility to chromium toxicity has not been reported. The most significant health effects associated with exposure to Cr(VI) involve the respiratory system and kidney. While effects on the respiratory system are unlikely to differ significantly with gender, the effect of gender on kidney toxicity is unknown.

5. DOSE-RESPONSE ASSESSMENTS

5.1. ORAL REFERENCE DOSE (RfD)

5.1.1. Choice of Principal Study and Critical Effect

Relatively few studies were located that addressed the oral toxicity of Cr(VI). One human study located in the literature, Zhang and Li (1987), reported on health effects in Chinese villagers who consumed drinking water from a well contaminated with hexavalent chromium from an alloy plant in Jinzhou. The Jinzhou area is heavily industrialized. In 1965, the well water in a nearby suburban area was found to be stained yellow, presumably because of chromium contamination from a mining operation which had begun operating in 1959. The mining operation was initially conducted in pilot scale, with a poor recovery rate for chromium (24.5%). Full-scale operation began in 1965. At this time, waste water was generated at a rate of

125 pounds/hour and contained concentrations of up to 105 mg/L hexavalent chromium. Waste water was deposited directly into a surface channel. Following sedimentation, the surface water concentration was still in excess of 20 mg/L. In addition to the surface water discharge, the mining operation generated hexavalent chromium-containing steam and a considerable amount of chromium-containing mine tailings. The tailings were stored in an open waste pile, containing approximately 300,000 pounds of waste, covering an area of 50 hectares. The waste pile constituted an additional source of ground water contamination through leaching and surface water runoff. In 1965, more than 28% of the area ground water samples were observed to be contaminated with chromium, with 54% of the samples contaminated at a concentration of 20 mg/L or greater. Ground water samples were found to be contaminated over an area of 10 square kilometers. In 1965, the material in the waste pile was found to contain an average of 1.55% hexavalent chromium. The soil in the vicinity of the waste pile was found to contain an average concentration of hexavalent chromium of 4,700 mg/kg, and 0.3 pounds/day of hexavalent chromium was estimated to leach into the ground water from the waste pile. In addition, irrigation water for the considerable agricultural operation in the vicinity of Jinzhou was contaminated with chromium at concentrations of 0.006-0.739 mg/L. However, concentrations in soil and produce in the agricultural areas were only slightly elevated above the controls.

In 1965, a study of 155 subjects exposed to drinking water at concentrations of approximately 20 mg/L of hexavalent chromium was conducted outside Jinzhou. Subjects were observed to have sores in the mouth, diarrhea, stomach ache, indigestion, and vomiting. Subjects were observed to have elevated white blood cell counts with respect to controls, as well as a higher per capita rate of cancers, including lung cancer and stomach cancer. Precise exposure concentrations, exposure durations, and confounding factors were not discussed, and this study does not provide a NOAEL for the observed effects. However, the study suggests that gastrointestinal effects may occur in humans following exposures to hexavalent chromium at levels of 20 ppm in drinking water (Zhang and Li, 1987).

Several animal studies addressing oral toxicity of hexavalent chromium were located in the literature. Anwar et al. (1961) exposed groups of female dogs (2/group) to up to 11.2 ppm Cr(VI) in drinking water for 4 years with no effect. MacKenzie et al. (1958) exposed groups of Sprague-Dawley rats (16-21/group) to up to 25 ppm Cr(VI) in drinking water for 1 year. No significant adverse effects were seen in any treatment group. Both the Mackenzie et al. (1958) and Anwar et al. (1961) studies are limited by the small number of animals/group and the lack of an observed effect at any dose level. While the MacKenzie et al. study has the additional limitation of being conducted for only 1 year, this study is considered to be more useful for risk assessment because of the identification of a NOAEL value and the larger number of animals per dose group that those available from Anwar et al. (1961).

Elbetieha and Al-Hamood (1997) reported adverse impacts on the male reproductive system and fertility, and reduced numbers of implantations and viable fetuses in pregnant females following exposures to hexavalent chromium. Information regarding the amount of water consumed by the animals was not provided, and this study is not considered useful for this risk assessment.

The National Toxicology Program (NTP 1996a,b, 1997) did not observe reproductive effects in either sex of BALB/C mice or Sprague-Dawley rats following oral exposures to 15 - 400 ppm Cr(VI) in the diet.

Junaid et al. (1996) and Kanojia et al. (1996) exposed female Swiss albino mice and female Swiss albino rats, respectively, to 250, 500, or 750 ppm potassium dichromate in drinking water to determine the potential embryotoxicity of hexavalent chromium during days 6-14 of gestation. The authors reported retarded fetal development and embryo- and fetotoxic effects including reduced fetal weight, reduced number of fetuses (live and dead) per dam, and higher incidences of stillbirths and postimplantation loss in the 500 and 750 ppm dosed mothers. Significantly reduced ossification in bones was also observed in the medium- and high-dose groups. Based on the body weight and the drinking water ingested by the animals in the 250 ppm dose group, the exposure levels in the 250 ppm groups can be identified as 67 mg/kg-day and 37 mg/kg-day in mice and rats, respectively.

The Junaid et al. (1996) and Kanojia et al. (1996) studies utilized doses approximately 10-fold higher than those used in Mackenzie et al (1958), but neither of the reproductive studies identified a clear NOAEL for the embryotoxic effects of hexavalent chromium. Based on the body weight and the drinking water ingested by the animals in the low-dose groups (250 ppm), the LOAELs of 67 mg/kg-day and 37 mg/kg-day can be identified from Junaid et al. (1996) and Kanojia et al. (1996) in mice and rats, respectively. Application of 10-fold uncertainty factor to extrapolate from LOAELs to NOAELs in these studies would generate NOAELs of 6.7 mg/kg-day and 3.7 mg/kg-day, respectively. These extrapolated NOAEL values are similar to, and support the use of, the NOAEL of 2.5 mg/kg-day identified from the study of MacKenzie et al. (1958) for development of the reference dose.

5.1.2. Method of Analysis

MacKenzie et al. exposed groups of eight male and eight female Sprague-Dawley rats to 0.45-11.2 ppm (0.45-11.2 mg/L) hexavalent chromium (as K_2CrO_4) for 1 year in drinking water. The control group (10/sex) received distilled water. A second experiment involved three groups of 12 male and 9 female rats. One group was given 25 ppm (25 mg/L) chromium (as K_2CrO_4), a second received 25 ppm chromium in the form of chromic chloride, and the controls again received distilled water. No significant adverse effects were seen in appearance, weight gain, or food consumption, and there were no pathologic changes in the blood or other tissues in any treatment group. The rats receiving 25 ppm of chromium (as K_2CrO_4) showed an approximate 20% reduction in water consumption. This dose corresponds to 2.5 mg Cr(VI)/kg/day based on actual body weight and water consumption data.

For rats treated with 0-11 ppm (in the diet), blood was examined monthly and tissues (livers, kidneys and femurs) were examined at 6 mo and 1 year. Spleens were also examined at 1 year. The 25 ppm groups (and corresponding controls) were examined similarly, except that no animals were killed at 6 mo. An abrupt rise in tissue chromium concentrations was noted in rats treated with greater than 5 ppm. The authors stated that “apparently, tissues can accumulate considerable quantities of chromium before pathological changes result.” In the 25 ppm

treatment groups, tissue concentrations of chromium were approximately 9 times higher for those treated with hexavalent chromium than for the trivalent group. Similar no-effect levels have been observed in dogs. Anwar et al. (1961) observed no significant effects in female dogs (2/dose group) given up to 11.2 ppm Cr(VI) (as K_2CrO_4) in drinking water for 4 years. The calculated doses were 0.012-0.31 mg/kg of Cr(VI).

5.1.3. RfD Derivation

No effects were reported at any dose level in the MacKenzie et al. study. The highest dose group (25 mg/L) was selected for derivation of the reference dose. Based on the body weight of the rat (0.35 kg) and the average daily drinking water consumption for the rat (0.035 l/day), this dose can be converted to give an adjusted NOAEL of 2.5 mg/kg-day.

The adjusted NOAEL is further modified by two 10-fold uncertainty factors to account for the expected interspecies and interhuman variability in lieu of specific data. An additional threefold uncertainty factor is applied to the adjusted NOAEL to compensate for the less-than-lifetime exposure duration in the MacKenzie et al. study. A threefold modifying factor is applied to address concerns raised by the study of Zhang and Li (1987). The total uncertainty factor applied to the adjusted NOAEL is 900. Application of the uncertainty factor to the adjusted NOAEL of 2.5 mg/kg-day gives the reference dose of 3×10^{-3} mg/kg-day.

5.2. INHALATION REFERENCE CONCENTRATION (RfC)

The inhalation reference concentration (RfC) is based on the assumption that thresholds exist for certain toxic effects such as cellular necrosis, but may not exist for other toxic effects such as carcinogenicity. In general, the RfC is an estimate (with uncertainty spanning perhaps an order of magnitude) of a daily exposure to the human population (including sensitive subgroups) that is likely to be without an appreciable risk of deleterious effects during a lifetime.

5.2.1. Choice of Principal Study and Critical Effect

Numerous studies have reported upper respiratory, lower respiratory, and kidney effects in humans and animals following exposures to hexavalent chromium. Of these endpoints, upper and lower respiratory effects appear to be the most sensitive and are discussed in greater detail below.

Three studies have focussed on nasal mucosal irritation, atrophy, and perforation following occupational exposures to chromic acid mists (Cohen et al., 1974; Lucas and Kramkowski, 1975; Lindberg and Hedenstierna, 1983). Of these, the study of Lindberg and Hedenstierna provides the most information on exposure levels and symptoms reported by exposed workers. Respiratory symptoms, lung function, and changes in nasal septum were studied in 104 workers (85 males, 19 females) exposed in chromeplating plants. Workers were interviewed using a standard questionnaire for the assessment of nose, throat, and chest

symptoms. Nasal inspections and pulmonary function testing were performed as part of the study.

The median exposure time for the entire group of exposed subjects (104) in the study was 4.5 years (0.1-36 years). Forty-three subjects exposed almost exclusively to chromic acid experienced a mean exposure time of 2.5 years (0.2-23.6 years). The subjects exposed almost exclusively to chromic acid were divided into a low-exposure group (8 hr TWA below 0.002 mg/m^3, N = 19) and high exposure group (8 hr TWA above 0.002 mg/m^3, N = 24). Exposure measurements using personal air samplers were performed for 84 subjects in the study on 13 different days. Exposure for the remaining 20 workers was assumed to be similar to that measured for workers in the same area. Nineteen office employees were used as controls for nose and throat symptoms. A group of 119 auto mechanics whose lung function had been evaluated by similar techniques was selected as controls for lung function measurements. Smoking habits of workers were evaluated as part of the study.

At mean exposures below 0.002 mg/m^3, 4/19 workers from the low-exposure group complained of subjective nasal symptoms. Atrophied nasal mucosa were reported in 4/19 subjects from this group and 11/19 had smeary and crusty septal mucosa, which was statistically higher than controls. No one exposed to levels below 0.001 mg/m^3 complained of subjective symptoms. At mean concentrations of 0.002 mg/m^3 or above, approximately one-third of the subjects had reddened, smeary, or crusty nasal mucosa. Atrophy was seen in 8/24 workers, which was significantly different from controls. Eight subjects had ulcerations in the nasal mucosa and five had perforations of the nasal septum. Atrophied nasal mucosa was not observed in any of the 19 controls, but smeary and crusty septal mucosa occurred in 5/19 controls.

Short-term effects on pulmonary function were evaluated by comparing results of tests taken on Monday and Thursday among exposed groups and controls. No significant changes were seen in the low-exposure group or control group. Nonsmokers in the high-exposure group experienced significant differences in pulmonary function measurements from the controls, but the results were within normal limits.

The authors concluded that 8-hour mean exposures to chromic acid above 0.002 mg/m^3 may cause a transient decrease in lung function, and that short-term exposures to greater than 0.02 mg/m^3 may cause septal ulceration and perforation. Based on the results of this study, a LOAEL of 0.002 mg/m^3 can be identified for incidence of nasal septum atrophy following exposure to chromic acid mists in chromeplating facilities. It should be noted that there are significant uncertainties related to the use of this LOAEL for development of an RfC for hexavalent chromium in the environment. There is considerable uncertainty with regard to the relevance of the nasal septum atrophy endpoint observed in the chromeplating industry to exposure to hexavalent chromium in the environment. The effects were observed in chromeplaters exposed to chromic acid mists near the plating baths. Environmental exposures would most likely occur through contact with hexavalent chromium dusts. An additional uncertainty is related to the determination of dose in the Lindberg and Hedenstierna study. Nasal septum atrophy in this study was related to time-weighted average (TWA) exposures to chromic acid. The most significant effects (nasal septum perforation) were observed in workers who

experienced peak excursions to levels considerably greater than the TWA. It is uncertain whether the peak excursion data or the TWA are more appropriate for the determination of dose in this study.

An alternative approach to development of the RfC is to focus on respiratory effects following inhalation of hexavalent chromium particulates. Two studies provide high quality data on lower respiratory effects following exposures to chromium particulates (Glaser et al., 1990; Glaser et al., 1985). Glaser et al. (1990) exposed 8-week-old male Wistar rats to sodium dichromate at 0.05 - 0.4 mg Cr(VI)/m^3 22 hr/day, 7days/week for 30-90 days. Chromium-induced effects occurred in a strong dose-dependent manner. The authors observed obstructive respiratory dyspnea and reduced body weight following subacute exposure at the higher dose levels. The mean white blood cell count was increased at all doses ($p < 0.05$) and was related to significant dose-dependent leukocytosis following subacute exposures. Mean lung weights were significantly increased at exposure levels of 0.1 mg/m^3 following both the subacute and subchronic exposures. Accumulation of macrophages was seen in all of the exposure groups and was postulated to be a chromium-specific irritation effect that accounted for the observed increases in lung weights. Focal inflammation was observed in the upper airways following the subchronic exposure, and albumin and LDH in BALF were increased following the exposure. The authors concluded that chromium inhalation induced pneumocyte toxicity and suggested that inflammation is essential for the induction of most chromium inhalation effects, and may influence the carcinogenicity of Cr(VI) compounds.

Glaser et al. (1985) exposed 5-week-old male Wistar rats to aerosols of sodium dichromate at concentrations ranging from 0.025 to 0.2 mg Cr(VI)/m^3, 22 hr/day in subacute (28 day) or subchronic (90 day) protocols. Chromium-induced effects occurred in a dose-dependent manner. Lung and spleen weights were significantly increased ($p < 0.005$) after both subacute and subchronic exposures at concentrations greater than 0.025 mg/m^3. Differences in the mean total serum immunoglobulin were also significant at exposures above 0.025 mg/m^3, while exposures to aerosol concentrations greater than 0.1 mg/m^3 resulted in depression of the immune system stimulation. The immune-stimulating effect of subchronic exposure was not reversed after 2 mo of fresh air regeneration. BAL cell counts were significantly decreased following subchronic exposure to levels above 0.025 mg/m^3 chromium. The number of lymphocytes and granulocytes showed a slight but significant increase in the lavage fluids of the subacute and subchronically exposed groups. At subacute exposure concentrations up to 0.05 mg/m^3, the phagocytic activity of the alveolar macrophages increased; however, subchronic exposure at 0.2 mg/m^3 decreased this function significantly. The spleen T-lymphocyte subpopulation was stimulated by subchronic exposure to 0.2 mg/m^3 chromium, and serum contents of triglycerides and phospholipids differed significantly from controls ($p < 0.05$) at this concentration.

Together, these studies provide useful information on chromium exposure-related impacts including lung and spleen weight, lactate dehydrogenase (LDH) in BALF, protein in BALF, and albumin in BALF. The cellular content of BALF is considered representative of initial pulmonary injury and chronic lung inflammation, which may lead to the onset of pulmonary fibrosis (Henderson, 1988). While these studies present dose-dependent results on sensitive indicators of lower respiratory toxicity, potential upper respiratory impacts resulting from the

exposures were not addressed. Glaser et al. (1990) states that the upper respiratory tract was examined, but these data are not reported. In light of the numerous reports of severe upper respiratory impacts following exposure to chromic acid in the occupational setting, the studies of Glaser et al. alone are not considered sufficient to support derivation of an RfC for chromium.

While the studies of Lindberg and Hedenstierna (1983) and Glaser et al. (1985, 1990) are independently considered insufficient for development of an RfC for hexavalent chromium, taken together these offer an approach for development of an RfC.

5.2.2. RfC Derivation

Lindberg and Hedenstierna (1983) will be used to support development of an RfC for upper respiratory effects of chromic acid mists and dissolved hexavalent chromium aerosols, and Glaser et al. (1985, 1990) will be used to support development of an RfC for lower respiratory effects from chromium particulates.

5.2.2.1. *Chromic Acid Mists and Dissolved Hexavalent Chromium Aerosols*

A LOAEL for nasal septum atrophy of 2 $\mu g/m^3$ chromic acid can be identified based on the results of Lindberg and Hedenstierna (1983). At TWA exposures greater than 2 $\mu g/m^3$, nasal septum ulceration and perforations occurred in addition to the atrophy reported at lower concentrations. The LOAEL is based on an 8-hour TWA occupational exposure. The LOAEL is adjusted to account for continuous exposure according to the following equation:

$$LOAEL_c = 2\ \mu g/m^3 \times (MVho/MVh) \times 5\ days/7\ days$$

where:

$LOAEL_c$ is the LOAEL for continuous exposure
MVho is the breathing volume for an 8-hour occupational exposure (10 m^3)
MVh is the breathing volume for a 24-hour continuous exposure (20 m^3)

The LOAEL of 2 $\mu g/m^3$ based on a TWA exposure to chromic acid is converted to a LOAEL for continuous exposure of 0.714 $\mu g/m^3$. An uncertainty factor of 3 is applied to the LOAEL to extrapolate from a subchronic to a chronic exposure, an uncertainty factor of 3 is applied to account for extrapolation from a LOAEL to a NOAEL, and an uncertainty factor of 10 is applied to the LOAEL to account for interhuman variation. The total uncertainty factor applied to the LOAEL is 90. Application of the uncertainty factor of 90 to the LOAEL of 0.714 $\mu g/m^3$ generates an RfC for upper respiratory effect of chromic acid mists and dissolved hexavalent chromium aerosols of 0.008 $\mu g/m^3$.

5.2.2.2. *Hexavalent Chromium Dusts*

Glaser et al. (1990) exposed male Wistar rats to 0.05-0.4 mg/m^3 sodium dichromate 22 hr/day, 7 days/week for 30 or 60 days, or 90 days with a 30-day recovery period. In Glaser et al. (1985), male Wistar rats were exposed to 0.025 - 0.2 mg/m^3 sodium dichromate 22 hr/day, 7 days/week for 28 or 90 days. Data were reported on numerous endpoints indicative of lung toxicity. One approach for development of an RfC using these data was offered by Malsch et al. (1994), who generated an inhalation RfC for chromium dusts using a benchmark concentration (BMC) approach. The Agency based its RfC derivation on this approach. After excluding exposures for periods of less than 90 days from the BMC analysis, Malsch et al. (1994) developed BMCs for lung weight, lactate dehydrogenase (LDH) in BALF, protein in BALF, albumin in BALF, and spleen weight. The Malsch et al. (1994) analysis defined the benchmark concentration as the 95% lower confidence limit on the dose corresponding to a 10% relative change in the endpoint compared to the control. Dose-effect data were adjusted to account for discontinuous exposure (22 hr/day) and the maximum likelihood model was used to fit continuous data to a polynomial mean response regression, yielding maximum likelihood estimates of 36 -78 µg/m^3 and BMCs of 16 - 67 µg/m^3. Malsch et al. (1994) applied dosimetric adjustments and uncertainty factors to the BMCs to determine a RfC based on the following equation:

$$RfC = \frac{BMC \times RDDR}{UF_A \times UF_F \times UF_H}$$

where:

RfC is the inhalation reference concentration
BMC is the benchmark concentration (lower 95% confidence limit on the dose corresponding to a 10% relative change in the endpoint compared to the control)
RDDR is the regional deposited dose ratio to account for pharmacokinetic differences between species
UF_A is a threefold uncertainty factor to account for pharmacodynamic differences not addressed by the RDDR
UF_F is a threefold uncertainty factor to account for extrapolating from subchronic to chronic exposures; and
UF_H is a 10-fold uncertainty factor to account for the variation in sensitivity among members of the human population

The RDDR factor is incorporated to account for differences in the deposition pattern of inhaled hexavalent chromium dusts in the respiratory tract of humans and the Wistar rat test animals (Jarabek et al., 1990). The RDDR of 2.1355 was taken from U.S. EPA (1990), based on the mass median aerodynamic diameter (0.28 µm for dose levels of 50-100 µg/m^3 and 0.39 for dose levels of 100-400 µg/m^3) and the geometric standard deviation (1.63 for dose levels of 50-100 µg/m^3 and 1.72 for dose levels of 100-400 µg/m^3) of the particulates reported in Glaser et al. (1990). A 3.16-fold uncertainty factor (midpoint between 1 and 10 on a log scale) was incorporated to account for the pharmacodynamic differences not accounted for by the RDDR. An additional 3.16-fold uncertainty factor was incorporated to account for the less-than-lifetime

exposure in Glaser et al. (1990), and a 10-fold uncertainty factor was applied to account for variation in the human population. A total uncertainty factor of 100 was applied to the BMC in addition to the RDDR.

Glaser et al. (1990) reported that LDH in BALF increased in a dose-dependent fashion from 50 to 400 $\mu g/m^3$ sodium dichromate, and this endpoint generated the lowest BMC (16 $\mu g/m^3$) and RfC (0.34 $\mu g/m^3$). LDH in BALF is considered the among the most sensitive indicators of potential lung toxicity (Henderson, 1984, 1985, 1988; Beck et al., 1982; Venet et al., 1985), as LDH is found extracellularly after cell damage and BALF is the closest site to the original lung injury. LDH in BALF may also reflect chronic lung inflammation, which may lead to pulmonary fibrosis through prevention of the normal repair of lung tissue (Henderson, 1988).

Several uncertainties must be addressed with regard to the BMC and RfC developed by Malsch et al. (1994). Potentially important endpoints including upper airway effects and potential renal or immunological toxicity were not addressed in the Glaser et al. (1985, 1990) studies and could not be included in the BMC analysis. While LDH in BALF resulted in the lowest BMC and RfC, several of the effects noted in Glaser et al. (1985, 1990) can be considered indicative of an inflammatory response, and might be equally suited to development of the RfC. In addition, the threefold uncertainty factor accounting for the use of a subchronic study may not be sufficiently protective for long-term effects. While the analysis acknowledged the importance of particle size and airway deposition in the development of the RDDR, the potential impact of different particle sizes in respiratory toxicity by hexavalent chromium particulates was not addressed.

Several of these uncertainties have been conservatively addressed in the analysis of Malsch et al. (1994). LDH in BALF generated the lowest estimate of the BMC from the effects noted by Glaser et al. (1985, 1990). This effect can be considered to be indicative of cell damage that occurs prior to fibrosis, as LDH appears in BALF following cell lysis. While other endpoints considered in the Malsch et al. (1994) analysis demonstrated a relatively better curve fit than LDH in BALF, the model generated a conservative fit in the data that is unlikely to overestimate the BMC, and the curve fit for LDH in BALF is considered to be acceptable. LDH in BALF as reported in Glaser et al. (1990) is considered to be an acceptable endpoint for development of an RfC for inhalation of hexavalent chromium particulates, and Malsch et al. (1994) used a reasonable approach for development of a BMC based on this endpoint.

The threefold uncertainty factor used to account for the subchronic study is insufficient for development of the RfC for inhalation of chromium particulates. Glaser et al. (1985) demonstrated that at the end of the 90-day exposure period, chromium was still accumulating in the lung tissue of the test animals, suggesting that lower long-term exposures might lead to accumulation of a critical concentration in the lung. Subchronic studies also may not adequately predict the presence of inflammatory effects from lower long-term exposures. The Agency has therefore determined that a 10-fold uncertainty factor accounting for the use of a subchronic study is more appropriate in this case for the development of the RfC for inhalation of chromium particulates.

Selection of a threefold uncertainty factor to account for the pharmacodynamic differences not accounted for by the RDDR, an additional 10-fold uncertainty factor to account for the less-than lifetime exposure in Glaser et al. (1990), and a 10-fold uncertainty factor to account for variation in the human population generates a total uncertainty factor of 300. Application of the total uncertainty factor of 300 and the RDDR of 2.1576 to the BMC generated by Malsch et al. (1994) based on LDH in BALF (Glaser et al., 1990) results in an RfC of 0.1 $\mu g/m^3$ for inhalation of hexavalent chromium particulates. The selected RfCs are 0.008 $\mu g/m^3$ for chromic acid mists and dissolved hexavalent chromium aerosols and 0.1 $\mu g/m^3$ for hexavalent chromium particulates.

5.3. CANCER ASSESSMENT

5.3.1. Summary

There are many epidemiologic studies demonstrating that hexavalent chromium (CrVI) is a potential human carcinogen, but few provide adequate exposure data for use in risk estimation. Mancuso (1975) provides limited but adequate information for this purpose, and Mancuso's data are used as the main database for estimating the carcinogenic potency of hexavalent chromium. Three foreign studies on ferrochromium plants were also considered for use in the potency calculations. From the quantitative risk assessment viewpoint, these studies are less adequate than the Mancuso study. For the Norwegian study (Langard et al., 1980), the exposure measurements were taken in 1975, while some workers could have been exposed to chromium as early as 1928, when the ambient dust levels were much higher than in later years. For the Swedish study (Axelsson et al., 1980), the chromium-exposed workers did not show a significant increase of lung cancer, and thus only the statistical upper bound of the response can be used in potency estimation. It is expected that the use of data from the Norwegian and Swedish studies would result in an overestimation of the true carcinogenic potency of hexavalent chromium. While a Russian study (Pokrovskaya and Shabynina, 1973) does not have the deficiencies of the other two foreign studies, the cohort in this study is not well defined and is not suitable for use in risk assessment.

Animal data from intratracheal studies were not used to estimate the carcinogenic potency of chromium by inhalation because there is limited pharmacokinetic information relating the distribution of chromium to lung tissues by inhalation and by intratracheal administration. This information is needed to reconcile the differences in dose distribution between these two exposure patterns. Furthermore, the physiological mechanism of dose distribution by intratracheal administration may depend (in a nonlinear fashion) on the dose levels used in the experiment, as evidenced by the observation that a single administration of sodium dichromate induced a carcinogenic response in Sprague-Dawley rats but failed to induce a response when the same weekly dose was given over 5 days (Steinhoff et al., 1983).

5.3.2. Dose-Response Data

The Mancuso (1975) study was based on a cohort of 332 white male workers who were employed in a chromate plant between 1931 (when the plant began to operate) and 1937, and who were followed to 1974. Mancuso reported lung cancer death rates by levels of exposure to soluble, insoluble, and total chromium concentrations. Because only lung cancer mortality for total chromium exposure was reported by age group, only the dose-response data for total chromium were used to estimate the carcinogenic potency of hexavalent chromium. The use of dose-response data for total chromium would result in an underestimation of the potency of hexavalent chromium. An additional uncertainty of the study was the assumption that the smoking habits of chromate workers were similar to those of the general white male population. This assumption may lead to an overestimation of the role of hexavalent chromium in lung cancer seen in the cohort.

Exposure information in the Mancuso study was derived from an industrial hygiene study of the plant conducted in 1949 (Bourne and Yee, 1950). In this study, TWAs of exposure to insoluble, soluble, and total chromium per cubic meter were calculated for each occupation and for each worker in every department. Using these data and company personnel records, Mancuso was able to calculate an estimate of exposure to soluble, insoluble, and total chromium by duration of exposure (in mg/m^3/years) for each member of the 1931-37 cohort. In 1949, after the industrial hygiene study had been conducted, the company initiated a comprehensive program designed to reduce employees' exposures and improve manufacturing efficiency. Until that time, however, the company had not undertaken any programs for the purpose of reducing employee exposure. It should be noted that Bourne and Yee (1950), who conducted the industrial hygiene survey in 1949, reported that "in order to meet price and quality competition, improvements in equipment and processes have been made periodically during the past 18 years, and it is the universal experience of industrial hygiene personnel that greater process efficiency is almost invariably associated with a more healthful working environment. Therefore, there seems little doubt that atmospheric contamination in the past was greater than in 1949." Nonetheless, no concerted effort was made to reduce employee exposure until late in 1949, and because this particular plant was a relatively modern one at the time of the survey, it is unlikely that improvements in efficiency over the period 1931 to 1949 would have reduced employee exposure to a great extent. Thus, Mancuso is considered to have utilized a reasonable approximation of what workers in the study cohort were exposed to during their entire working history. Exposure in the cohort may be slightly underestimated because of the likelihood that a greater proportion of the "total exposure" was contributed prior to 1949 than after 1949. The effects of underestimating the exposure concentration, as well as the effects of other uncertainties on the estimation of potency, are addressed in Section 6.

5.3.3. Dose Conversion

Table 3, which is taken from Mancuso (1975), presents age-specific lung cancer deaths, corresponding person-years, and range of exposures to total chromium. To estimate the lifetime cancer risk due to exposure to chromium, it is assumed that an exposure, D (mg/m^3/years), as presented in Table 3, is equivalent to the continuous exposure d ($\mu g/m^3$) calculated by

$$d = \frac{D}{fL_e} \times \frac{8}{24} \times \frac{240}{365} \times 10^3 \ \mu g/m^3$$

where L_e is the midrange in each age category, f is the fraction of time in age exposed, and 8/24 and 240/365 are the fractions of a day and year, respectively, that a worker spent at the plant. For instance, if D = 8 mg/m^3/years, L_e = 60, and f = 0.65, then d = 44.96 µg/m^3. The assumption of f = 0.65 implies that the cohort exposure to chromium began at approximately age 20. The assumption is that the particular exposure pattern (unknown) leading to the cancer mortality rates as observed is equivalent to the continuous constant exposure starting from the age when exposure began. This assumption may or may not be realistic. However, it would be less realistic to make a different assumption concerning the exposure pattern when the exposure estimates provided in Mancuso (1975) were determined using the weighted average of the duration of exposure for each respective job the worker had.

Since the person-year in each category presented in Table 2 is very small, the exposure categories are combined as shown in Table 4 to increase statistical stability. The last column of Table 4 is given for the purpose of identifying which exposure categories in Table 2 are combined. The midrange of age and exposure concentration is used in Table 4. Data in this table are used to estimate the lifetime cancer risk due to chromium exposure.

5.3.4. Extrapolation Method

It has been widely recognized (e.g., Doll, 1971) that the age-specific incidence curve tends to be linear on doubly logarithmic graphs, or equivalently, the age-specific incidence follows the mathematical form:

$$I(T) = bT^{k-l}$$

where b and k are parameters that may be related to other factors such as dose, and T may be one of the following three cases:

1. T is age when cancer is observed,

2. T is the time from the first exposure to observed cancer, or

3. T is the time from exposure to cancer minus the minimum time for a cancer to be clinically recognized.

Table 4. Combined age-specific lung cancer death rates and total chromium exposure (in μg/m^3)

Age	Concentration (μg/m^3)	Deaths	Person-years	Background rate[b]	Exposure range
50	5.66	3	1,345	6.05×10^{-4}	≤1.99
50	25.27	6	931	6.05×10^{-4}	2.0-5.99
50	46.83	6	299	6.05×10^{-4}	6.0-7.99
60	4.68	4	1,063	1.44×10^{-3}	≤1.99
60	20.79	5	712	1.44×10^{-3}	2.0-5.99
60	39.08	5	211	1.44×10^{-3}	6.0-7.99
70	4.41	2	401	1.57×10^{-3}	≤1.99
70	21.29	4	345	1.57×10^{-3}	2.0-7.99

[a]The midrange of each exposure category in Table 2 is first converted to μg/m^3 by using f = 0.65 in the formula described in the section "data available for potency calculations." The concentrations presented in this table are the averages of several exposure categories weighted by corresponding person-years.

[b]Background rate is estimated from 1964 U.S. Vital Statistics. The year 1964 is selected because it is estimated that a large proportion of lung cancer deaths occurred during that year.

This model has been shown to arise from the somatic mutation hypothesis of carcinogenesis (Armitage and Doll, 1954; Whittemore, 1978; Whittemore and Keller, 1978). It has also been shown to arise from the epigenetic hypothesis when the reversible cellular change is programmed to occur randomly (Watson, 1977). These authors and many others have used this model to interpret and/or estimate potency from human data.

Since the data that could be used for risk estimation are limited, a simple model that fits the data should be used. Therefore, the observed age-specific incidence is assumed to follow the model

$$I(t,d) = B(t) + h(t,d)$$

where B(t) is the background rate at age t and $h(t,d) = Q(d)\, t^{k-1}$ with $Q(d) = q_1 d + q_2 d^2$, a function of dose d.

Once the parameters q_1, q_2, and k are estimated, the lifetime cancer risk associated with an exposure d by age t, taking into account the competing risk, can be calculated by

t s

$$P(t,d) = \int_0^s h(s,d)\exp\{-[\int_0^s h(y,d)dy + A(s)]\}ds$$

where exp[-A(s)] is the probability of surviving to age s and h(t,d) = I(t,d) - B(t) is the age-specific incidence after adjusting the background rate.

To estimate the parameters in h(t,d) we assume, as is usually done, that the number of lung cancer deaths, X, at age t, follows the Poisson distribution with the expected value

$$E(X) = N \times [B + Q(d)\, t^{k-1}]$$

where N is the person-year associated with X, B is the background rate at age t, and $Q(d) = q_1 d + q_2 d^2$.

Using the BMDP computer program P3R and the theory relating the maximum likelihood and nonlinear least square estimation by Jennrich and Moore (1975), the parameters q_l, q_2, and k are estimated by the method of maximum likelihood as $q_l = 1.11 \times 10^{-7}$, $q_2 = 1.84 \times 10^{-9}$, and k = 2.915; the corresponding standard deviations are respectively 7.8×10^{-7}, 1.2×10^{-8}, and 1.7.

Thus, the age-specific cancer death incidence at age t due to chromium exposure d $\mu g/m^3$ is given by

$$h(t,d) = Q(d)\, t^{l.915}$$

where

$$Q(d) = 1.11 \times 10^{-7} d + 1.84 \times 10^{-9} d^2$$

The model fits the data well, as can be seen from the goodness-of-fit statistic

$$x^2 = \sum (0\text{-}E)^2/E = 1.60$$

which has, asymptotically, a chi-square distribution with 5 degrees of freedom under the model specified. The observed and predicted values used in calculating x^2 are (3, 2.5), (6, 7.2), (6, 5.1), (4, 3.1), (5, 6.7), (5, 4.1), (2, 1.4), and (4, 4.3).

Taking into account the competing risk, the lifetime probability of lung cancer death due to exposure to chromium d $\mu g/m^3$ is given by

$$P(L,d) = \int_0^L h(t,d)\exp\{-[(Q(d)/2.915)\, t^{2.915} + A(t)]\}dt$$

where L is the maximum human lifetime and is mathematically equivalent to infinity, since the probability of surviving beyond L is 0. At low doses approximately,

$$P(L,d) = d \times P(L,l)$$

where P(L,l) is the lifetime cancer risk due to exposure to 1 $\mu g/m^3$ of chromium. The unit risk, P(L,l), has been adopted by the EPA as an indicator of the carcinogenic potency of a chemical compound.

5.3.5. Inhalation Unit Risk

To calculate the unit risk, P(L,1) it is necessary to know exp[-A(t)], the probability of surviving to age t. Since this probability can only be estimated, it is assumed that the survival probability is constant over a 5-year interval, as provided in the U.S. Vital Statistics.

Using this approximation and by integrating the formula P(L,1) we have

$$P(L,1) = \sum[\exp(-3.87 \times 10^{-8}\, t_{i-1}^{2.9l5}) - \exp(-3.87 \times 10^{-8}\, t_i^{2.9l5})] \times P_i$$

$$= 1.16 \times 10^{-2}/\mu g/m^3$$

where (t_{i-l}, t_i) is a 5-year interval and P_i is the probability of survival up to the age t_{i-l}. P_i is assumed to be a constant over the interval and is estimated from the 1975 U.S. Vital Statistics.

As a crude approximation, the carcinogenic potency of chromium can be also be calculated by $B = (R-1) \times P_o/d$, where $P_o = 0.036$ is the estimated lung cancer mortality rate for the U.S. population, R is the relative risk of the lung cancer deaths in the cohort, and d is the "standardized" lifetime dose concentration to which the workers were assumed to be exposed. This approach is used by EPA to calculate carcinogenic potency when the only data available are the relative risk estimate and an average exposure concentration.

For the Mancuso (1975) data, the relative risk R and the "standardized" dose d are estimated respectively to be R = 7.2 and d = 15.5 $\mu g/m^3$. They are calculated by combining the relative risks and dose concentrations in each of the age-exposure categories, weighted by the relative magnitude of person-years, as shown in Table 4.

Therefore, the carcinogenic potency of hexavalent chromium (CrVI) is estimated to be

$$B = (7.2\text{-}1) \times 0.036/15.5 = 1.4 \times 10^{-2}/\mu g/m^3$$

This crude estimate is only slightly higher than the previous estimate, $1.2 \times 10^{-2}/\mu g/m^3$.

6. MAJOR CONCLUSIONS IN THE CHARACTERIZATION OF HAZARD AND DOSE RESPONSE

6.1. HUMAN HAZARD POTENTIAL

Chromium is a naturally occurring element present in rocks, soils, plants, animals, and volcanic emissions. Chromium may exist in several chemical forms and valence states in the

environment. The most commonly occurring valence states are chromium metal (0), trivalent Cr(III), and hexavalent Cr(VI). The primary sources of hexavalent chromium in the environment are most likely chromate chemicals used as rust inhibitors in cooling towers and emitted as mists, particulate matter emitted during manufacture and use of metal chromates, and chromic acid mist from the chromeplating industry. Hexavalent chromium in the atmosphere may react with dust particles or other pollutants to form trivalent chromium, or may be removed from air by atmospheric fallout and precipitation. Hexavalent chromium may exist in aquatic media as watersoluble complex anions and may persist in water. Hexavalent chromium may also may react with organic matter or other reducing agents to form trivalent chromium. Hexavalent chromium in soil tends to be reduced to trivalent chromium by organic matter.

Cr(III) potentiates the action of insulin in peripheral tissue and is essential for animals and human beings. Adults in the United States are estimated to ingest approximately 60 µg/day of chromium from food (ATSDR, 1993). The national Research Council has identified an estimated safe and adequate daily dietary intake (ESADDI) for chromium of 50-200 µg/d (NRC, 1989), corresponding to 0.71-2.9 µg/kg/day for a 70 kg adult. FDA has selected a Reference Daily Intake for chromium of 120 µg/d (DHHS, 1995).

The bioavailability of chromium may be the single most important factor determining the toxicity of a specific chromium source (O'Flaherty, 1996). Ingested hexavalent chromium is efficiently reduced to the trivalent form in the gastrointestinal tract. Gastrointestinal absorption of Cr(VI) occurs with greater efficiency than absorption of Cr(III), though absorption of ingested hexavalent chromium is estimated to be less than 5%. Following inhalation exposure, chromium may be absorbed into the systemic circulation, transferred to the gastrointestinal tract by mucociliary action, or remain in the lung. A number of factors can influence the absorption of chromium following inhalation, including the size, oxidation state, and solubility of the chromium particles; the activity of alveolar macrophages; and the interaction of chromium with biomolecules following deposition in the lung. Inhaled hexavalent chromium can be reduced to the trivalent form by ascorbate and glutathione. Absorption of inhaled chromium following occupational exposure has been demonstrated by the measurement of chromium in the serum and urine and hair of workers in the chromium industry. Water-soluble hexavalent chromium has been shown to be absorbed rapidly by inhalation in rats.

A significant amount of absorbed chromium is taken up in the bone, liver, kidney, and spleen. Hexavalent chromium readily crosses cell membranes through the phosphate and sulfate anion-exchange carrier pathway. Cr(III) compounds may cross cell membranes, but only with very low efficiency. Cr(VI) readily passes through cell membranes and produces a number of potentially mutagenic DNA lesions upon intracellular reduction to Cr(III). Hexavalent chromium is mutagenic in bacterial assays, yeasts, and V79 cells, and transforms both primary cells and cell lines.

Results of occupational epidemiologic studies of chromium-exposed workers across investigators and study populations consistently demonstrate that chromium is carcinogenic by the inhalation route of exposure. While data from these studies could be used to suggest that total chromium is carcinogenic by inhalation, animal data support the human carcinogenicity data

only on hexavalent chromium. Hexavalent chromium compounds have been shown to produce the following tumor types in animal assays: intramuscular injection site tumors in rats and mice, intrapleural implant site tumors in rats, intrabronchial implantation site tumors in rats, and subcutaneous injection site sarcomas in rats. Workers are exposed to both Cr(III) and Cr(VI) compounds. Because only Cr(VI) has been found to be carcinogenic in animal studies, however, it was concluded that only Cr(VI) should be classified as a human carcinogen.

At present, the carcinogenicity of hexavalent chromium by the oral route of exposure cannot be determined because of a lack of sufficient epidemiological or toxicological data. One study of miners in Ontario suggested that exposure to chromium may have been associated with stomach cancer, but other human and animal studies have not reported similar effects.

A number of epidemiological studies of workers in chromium production facilities have demonstrated an association between inhalation of Cr(VI) and upper respiratory irritation and atrophy, lower respiratory effects, and renal effects. There is significant uncertainty regarding the relevance of occupational exposures to chromic acid mists to environmental exposures to hexavalent chromium particulates, as well as the role of direct contact between chromium-contaminated hands and nasal passages in the studies reporting nasal irritation, atrophy, and nasal septum perforation in the occupational setting. Animal studies have reported a variety of effects including perforation of the nasal septum, necrosis, atrophy and hyperplasia of the bronchial epithelium, bronchiolization of the alveoli, alveolar proteinosis, changes in lung weight, lactate dehydrogenase in BALF, albumin in BALF, changes in tracheal and submandibular lymph nodes, atrophy of the spleen and liver, and ulcerations in the stomach and intestinal mucosa following exposures to Cr(VI) compounds by inhalation (Steffee and Baetjer, 1965; Nettesheim et al., 1971; Glaser et al., 1985; Glaser et al., 1990).

Little data exist regarding health effects resulting from ingestion of hexavalent chromium. A single cross-sectional study was located that reported effects in humans resulting from ingestion of chromium-contaminated well water. Residents of a village in China were reported to have experienced oral ulcers, diarrhea, abdominal pain, indigestion, vomiting, leukocytosis, and presence of immature neutrophils. Other reports of toxic effects of Cr(VI) in humans are limited to case reports from accidental poisonings. With the exception of increased body burden of chromium, no significant adverse effects have been observed in animal studies following ingestion of chromium.

High oral doses of hexavalent chromium compounds have been reported to cause reproductive and developmental toxicity in mice, including decreased fetal weight, increased resorptions, and increased abnormalities. A recent study in mice and rats determined that hexavalent chromium is not a reproductive toxicant in either sex.

Chromium is one of the most common contact sensitizers in industrialized countries, and allergic contact dermatitis is associated with occupational exposures to numerous materials and processes, including chromeplating baths, chrome colors and dyes, cement, tanning agents, and wood preservatives.

6.2. DOSE RESPONSE

The data of Mancuso (1975) were used to generate the unit risk for inhalation of hexavalent chromium of $1.2 \times 10^{-2}/\mu g/m^3$. A recent follow-up study (Mancuso, 1997) is supportive of the conclusions of Mancuso (1975); however, several important uncertainties in the potency estimate result from the use of the Mancuso data for the dose-response estimation.

The risk of hexavalent chromium is estimated on the basis of the total chromium obtained from all the soluble and insoluble chromium to which workers were exposed. Since there are likely differences between the chromium compounds to which workers were exposed, the potency of hexavalent chromium compounds may be underestimated. Bourne and Yee (1950) reported that the ratios of Cr(III) to Cr(VI) concentrations in the airborne dust in nine major departments in the plant in which the Mancuso cohort worked ranged from 1 to 3, except for two departments where the ratios were 6 for the lime and ash operation and 52 for the ore preparation. Excluding the ore operation, the maximum ratio of trivalent chromium to hexavalent chromium is 6, and thus the underestimation of the risk for hexavalent chromium is unlikely to be greater than sevenfold.

Use of the hygiene data collected in 1949 may result in a slight underestimation of the levels of exposure workers experienced between 1931 and 1937. However, because the plant was relatively modern in the 1930s, the underestimation is unlikely to be large. If an underestimation of 2 times were assumed, then the unit risk would be reduced from $1.2 \times 10^{-2}/\mu g/m^3$ to $6 \times 10^{-3}/\mu g/m^3$.

The risk presented in this report may be somewhat overestimated as a result of the assumption that smoking habits of chromate workers were similar to those of the general white male population. It is generally accepted that the proportion of smokers is higher for industrial workers (thus the higher background incidence rates) than for the general population. For example, the background age-specific rate of lung cancer at ages 50, 60, and 70 could be 40% greater than that presented in Table 4 should it be assumed that 80% of the chromate workers in the Mancuso study were ever-smokers (individuals who smoke at least 100 cigarettes during their lifetimes) and only 50% of the general white male population were ever-smokers. For example, if the background rate of lung cancer mortality (due to smoking) for the cohort in Table 4 is increased by 40%, then the corresponding unit risk would be reduced by about 25%, or from $1.2 \times 10^{-2}/\mu g/m^3$ to $8.7 \times 10^{-3}/\mu g/m^3$.

Relatively few studies in the literature address the oral toxicity of Cr(VI). The Zhang and Li (1987) human study reported on health effects in 155 Chinese villagers who consumed drinking water contaminated with hexavalent chromium at 20 ppm. Only one exposure level was included and the study did not indicate whether the drinking water was contaminated with other materials in addition to hexavalent chromium. The study also did not address whether potential airborne exposures to hexavalent chromium from the plant or other confounding factors may have contributed to observed effects. The exposure period was unknown, and the study could not provide a NOAEL for the observed effects. However, the study of Zhang and Li suggests that

gastrointestinal effects in humans may occur at an exposure level of 20 ppm of hexavalent chromium in drinking water.

Two studies have reported fetotoxic and developmental effects of Cr(VI) in mice and rats at exposure levels of 250 - 700 ppm in drinking water (Junaid et al., 1996; Kanojia et al., 1996). While neither of these studies provide clear NOAEL values, LOAELs for fetotoxicity can be used to extrapolate to NOAELs of 6.7 mg/kg-day and 3.7 mg/kg-day in mice and rats, respectively.

Both the MacKenzie et al. (1958) and Anwar et al. (1961) animal studies are limited by a small number of animals/group and a lack of an observed effect at any dose level. The MacKenzie et al. study has the additional limitation of being conducted for less than a lifetime. However, the MacKenzie study was considered to be most suitable for the dose-response assessment for ingested chromium and generated an adjusted NOAEL of 2.5 mg/kg-day.

The adjusted NOAEL from the MacKenzie study was modified by two 10-fold uncertainty factors to account for the expected interspecies and interhuman variability in lieu of specific data. An additional threefold uncertainty factor is applied to the adjusted NOAEL to compensate for the less-than-lifetime exposure duration in the Mackenzie study. A modifying factor of 3 is applied to account for uncertainties resulting from study of Zhang and Li. The total uncertainty factor applied to the adjusted NOAEL is 900, yielding an RfD of 3×10^{-3} mg/kg-day. Confidence in the oral reference dose is low. Confidence in the chosen study is low because of the small number of animals tested, the small number of parameters measured, and the lack of toxic effect at the highest dose tested. Confidence in the database is low because the supporting studies are of equally low quality, because of concerns raised by the study of Zhang and Li, and because of the lack of information on teratogenic endpoints. Low confidence in the RfD follows.

Two RfCs have been generated for hexavalent chromium. The RfC for chromic acid mists and dissolved hexavalent chromium aerosols is based on a study of workers in a chromium plating facility (Lindberg and Hedenstierna, 1983). The occurrence of nasal mucosal atrophy in the Lindberg and Hedenstierna study is consistent with previous reports that exposure to chromium acid mists is associated with ulceration of the mucous membranes and perforation of the cartilaginous portions of the nasal septum (Hamilton and Hardy, 1974).

Several uncertainties result from the exposure characterization in the Lindberg and Hedenstierna (1983) study. While nasal mucosal atrophy has been consistently reported following occupational exposure to chromic acid mists, it is uncertain whether these exposures are relevant to exposures to hexavalent chromium particulates in the environment. The LOAEL for this study is based on an 8-hour TWA concentration. However, it is possible that the exposure response may be better explained by the peak exposure rather than the TWA dose. The authors acknowledge that nasal septum ulcerations and perforations did not correlate with mean exposure concentrations in the 2-20 $\mu g/m^3$ group and observed that damage to the nasal septum correlated better with short-term peak exposure than with 8-hour mean exposures. The report does not provide sufficient detail on peak exposures to resolve this issue unequivocally, and in the absence of additional data, the RfC was generated based on the TWA exposure. The RfC

based on Lindberg and Hedenstierna (1983) is applicable only to chromic acid mists and dissolved hexavalent chromium aerosols.

Confidence in the RfC for chromic acid mists and dissolved hexavalent chromium aerosols is low. Confidence in the chosen study is low because uncertainties regarding the exposure characterization and the role of direct contact for the critical effect. Confidence in the database is low because the supporting studies are equally uncertain with regard to the exposure characterization. Low confidence in the RfC follows.

The RfC for hexavalent chromium particulates was developed based on Glaser et al. (1985, 1990). Several uncertainties are associated with the use of these studies for development of the RfC. Glaser et al. (1985, 1990) did not provide details of upper respiratory, reproductive, or renal effects resulting from the exposures, and did not include chromic acid mists or dissolved hexavalent chromium aerosols in their study. This uncertainty has been addressed by limiting the RfC developed on the basis of Glaser et al. (1985, 1990) to lower respiratory effects from inhalation of hexavalent chromium dusts. Uncertainty results from the use of a subchronic study for development of the chronic RfC. This uncertainty was addressed by the use of a 10-fold uncertainty factor to account for potential chronic effects. It is uncertain which of the endpoints reported in Glaser et al. (1985, 1990) is most appropriate for development of the BMC. LDH in BALF was used for development of the RfC, as this endpoint is considered to be a sensitive indicator of toxicity and provides the most conservative estimate of the BMC based on the Glaser et al. (1985, 1990) data. Uncertainty in the dose-response curve for LDH in BALF was addressed through the use of a conservative model fit of the data.

Confidence in the RfC for hexavalent chromium particulates is medium. Confidence in the chosen study is medium because of uncertainties regarding upper respiratory, reproductive, and renal effects resulting from the exposures. Medium confidence in the RfC follows.

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APPENDIX A. EXTERNAL PEER REVIEW— SUMMARY OF COMMENTS AND DISPOSITION

The support document and IRIS summary for hexavalent chromium have undergone both internal peer review performed by scientists within EPA and a more formal external peer review performed by scientists performed accordance with EPA guidance on peer review (U.S. EPA, 1992). Comments made by the internal reviewers were addressed prior to submitting the documents for external peer review and are not part of this appendix. The external peer reviewers were tasked with providing written answers to general questions on the overall assessment and on chemical-specific questions in areas of scientific controversy or uncertainty. A summary of significant comments made by the external reviewers and EPA's response to these comments follows.

Comments on General Questions for IRIS Peer Reviewers

1. *Are you aware of any other data/studies that are relevant (i.e., useful for the hazard identification or dose-response assessment) for the assessment of the adverse health effects, both cancer and noncancer, of this chemical?*

A. Comment: Since Mancuso has recently updated his cohort and since the information is already referenced in the IRIS and the Toxicological Review document, I believe the cancer slope factors should be recalculated from the total chromium data and the Cr(VI) data.

Response to Comment: The Agency agrees that the cancer slope factor should be recalculated from the updated cohort. The Agency is unable to do so in a time frame consistent with the IRIS Pilot process, and will take up revision of the slope factor when possible.

B. Comment: In calculating the oral reference dose the authors used data from a rodent study by MacKenzie et al. (1958) and discounted data from a human study by Zhang et al. I am concerned that the NOAEL in the animal study exceeded the level reported in the Chinese study (20 ppm) that caused serious illness in humans.

Response to Comment: The study of Zhang and Li (1987) raises concerns for human gastrointestinal effects at high dose levels in drinking water; however, the exposure data in this study are limited, the duration of exposure is unknown, the presence of confounding factors is not addressed, and a NOAEL could not be identified from the study. While these uncertainties preclude the use of this study for risk assessment, a threefold modifying factor has been applied to the NOAEL derived from the study of MacKenzie et al. (1958) to address the concerns raised by the study of Zhang and Li (1987).

C. Comment: Several new reports have been found showing significant embryotoxic and fetotoxic damage due to exposure of rats and mice to high doses of Cr(VI) or Cr(III) in drinking water. I don't feel confident rederiving an RfD based on this data, and the doses are clearly very high. However, derivation of an RfD based on an observed toxicological effect appears to be preferable to an RfD based on a NOAEL.

Response to Comment: The reports have been added to the reproductive/developmental studies section of the toxicological review document and have been considered in the development of the RfD for Cr(VI). The new reports do not provide a clear NOAEL for the fetotoxic and embryotoxic effects. Extrapolated LOAELs derived from the studies of Junaid et al. (1996) and Kanojia et al. (1996) are similar to and support the use of the NOAEL identified in the study of MacKenzie et al. (1958) for development of the RfD.

D. Comment: The documents correctly state that Cr(VI) gets transformed to Cr(III) in vivo, but they skirt the issue of whether or not a Cr(VI) study is really a study of in vivo exposure to Cr(III).

Response to Comment: Given the rapid reduction of Cr(VI) to Cr(III) in vivo, it is relevant to consider whether environmental exposures to Cr(VI) or administration of Cr(VI) in controlled animal experiments is essentially identical to environmental exposures to Cr(III) or administration of Cr(III) in controlled experiments. While considerably more data are available for Cr(VI) than for Cr(III), it appears at present that exposures to Cr(VI) have considerably different outcomes than exposures to Cr(III). The Agency has prepared the toxicological summaries and IRIS files for Cr(VI) and Cr(III) from this perspective.

2. *For the RfD and RfC, has the most appropriate critical effect been chosen (i.e., that adverse effect appearing first in a dose-response continuum)? For the cancer assessment, are the tumors observed biologically significant? Relevant to human health? Points relevant to this determination include whether or not the choice follows from the dose-response assessment, whether the effect is considered adverse, and if the effect (including tumors observed in the cancer assessment) and the species in which it is observed is a valid model for humans.*

A. Comment: The descriptions of the study used to develop the RfC for chromic acid mist were confusing. While I agree that the LOAEL is 0.002 mg/m^3, I question the uncertainty factor of 3 to extrapolate from subchronic to chronic exposure. Virtually all the workers in the high dose level in the Lindberg cohort had significant effects even though the duration of exposure for workers with nasal ulcerations ranged from 5 mo to more than 10 years.

Response to Comment: The descriptions of the study of Lindberg and Hedenstierna (1983) have been improved. The experience of workers at the high dose level with significant effects over a subchronic exposure period does not eliminate the possibility that similar effects could occur at considerably lower doses over a chronic exposure period. In order to account for this uncertainty, the Agency favors use of a threefold factor to extrapolate from the subchronic to chronic exposures.

B. Comment: The authors argue that the threefold uncertainty factor used by Malsch et al. is insufficient. I disagree. In the Glasser paper the concentration in the lung appears to be approaching a maximum at 90 days. The authors also suggest that inflammatory effects from lower long-term exposures may occur. In my experience inflammation is an early symptom and

in some cases even regresses in the presence of continuing exposure. Thus, I would use Malsch's calculations as published yielding a RfC of 0.34 mg/m^3.

Response to Comment: Information from Glaser (1985) shows that chromium is still accumulating in the lung and kidney at the end of the 90-day exposure period and there is insufficient information to determine how well subchronic studies predict chronic inflammation. The Agency supports the use of a 10-fold uncertainty factor to extrapolate from a subchronic to a chronic study.

C. Comment: The additional uncertainty factor of 3 (for the RfD) to compensate for less than lifetime exposure duration in the MacKenzie et al. study can be questioned. Why not apply a 10-fold uncertainty factor for this purpose, based also on the relatively low number of animals used in the study?

Response to Comment: The use of the MacKenzie et al. study for development of the RfD is considered to be conservative, given the lack of an observed effect in the animals. Even in light of the relatively low number of animals in the study, the threefold uncertainty factor is considered by the Agency to be sufficient for development of the RfD.

3. *Have the noncancer and cancer assessments been based on the most appropriate* studies? *These studies should present the critical effect/cancer (tumors or appropriate precursor) in the clearest dose-response relationship. If not, what other study (or studies) should be chosen and why?*

A. Comment: A level of 0.002 mg/m^3 in the Lindberg and Hedenstierna study was chosen as the LOAEL for the RfC. This level caused significant symptoms, including atrophied nasal mucosa, in workers exposed, whereas no symptoms were seen at a level of 0.001 mg/m^3. Therefore, why not use this lower level as a NOAEL?

Response to Comment: Although no subjective irritation occurred in the subgroup exposed at 0.001 mg/m^3, the distribution of the four cases of atrophy in this group was not provided, which precluded designation of a NOAEL at 0.001 mg/m^3.

B. Comment: The RDDR is based on aerodynamic diameters of particles used in the animal studies; however, it would be helpful to also know something about ambient particle sizes containing chromium such that a more appropriate dosimetric adjustment across species using differences in particle deposition between rodents and humans can be applied.

Response to Comment: Data on ambient particle sizes containing chromium are necessarily site specific, and will vary depending on the nature of the contaminated media and exposure setting. While it might be of interest to determine the ambient particle size distribution in order to develop a site-specific RDDR, these data cannot be practically incorporated into the RDDR in order to account for all of the possible ambient exposures. The assessment has utilized the data in the Agency's guidance for development of RfCs for this purpose.

C. Comment: It appears that the endpoint albumin in bronchoalveolar lavage fluid showed even greater effects than LDH, and I wonder why this endpoint was not selected for the benchmark approach.

Response to Comment: LDH in BALF provided a BMD approximately one-half that of albumin in BALF (Malsch et al., 1994). In order to be conservative and focus on the critical effect, LDH in BALF was chosen as the endpoint for development of the RfC.

D. Comment: The fact that chromium was still accumulating in lung tissue at the end of a 90-day exposure does not suggest that lower long-term exposures will lead to accumulation of a critical concentration in the lung. This depends very much on the clearance kinetics, and at low concentrations lung levels will reach an equilibrium that is lower than that achieved at higher concentrations.

Response to Comment: The Agency acknowledges the possibility that at low concentrations lung levels will reach an equilibrium which is lower than that achieved at higher concentrations. However, in the absence of data, it cannot be demonstrated that chronic exposures will not lead to accumulation of a critical concentration in the lung. In order to conservatively reflect the uncertainty on this issue, the Agency has utilized a 10-fold uncertainty factor to account for less-than lifetime exposure.

4. *Studies included in the RfD and RfC under the heading "Supporting/Additional studies" are meant to lend scientific justification for the designation of critical effect by including any relevant pathogenesis in humans, any applicable mechanistic information, any evidence corroborative of the critical effect, or to establish the comprehensiveness of the database with respect to various endpoints (such as reproductive/developmental toxicity studies). Should other studies be included under the "Supporting/Additional" category? Should some studies be removed?*

A. Comment: The new data on reproductive toxicity of chromium in the drinking water needs to be carefully compared to the NTP study in which rats and mice were fed potassium chromate in the diet. The form of chromium and route of exposure are clearly of paramount importance.

Response to Comment: A discussion comparing and contrasting the results of the NTP studies and the new reports has been added to the reproductive/developmental studies section of the toxicological review document.

B. Comment: Some of the statements related to the genotoxic effects of hexavalent chromium are either inaccurate or misleading.

Response to Comment: The recommended modifications to this section have been made.

5. *For the noncancer assessments, are there other data that should be considered in developing the uncertainty factors of the modifying factor? Do you consider that the data support the use of different (default) values than those proposed?*

A. Comment: No comment received.

6. *Do the confidence statements and weight-of-evidence statements present a clear rationale and accurately reflect the utility of the studies chosen, the relevancy of the effects (cancer and non-cancer) to humans, and the comprehensiveness of the database? Do these statements make sufficiently apparent all the underlying assumptions and limitations of these assessments? If not, what needs to be added?*

A. Comment: Yes.

Comments on Chemical-Specific Questions

1. *Are the conclusions of Zahid et al. regarding potential reproductive toxicity of Cr(III) in any way countered by the results of the NTP study?*

A. Comment: The Cr(VI) document does a superficial job of communicating the deficiencies of the Zahid study.

Response to Comment: Additional information has been provided in the reproductive/developmental effects section to address this concern.

2. *Should separate RfCs be generated for chromic acid mists and particulates of hexavalent chromium?*

A. Comment: Yes.

B. Comment: Yes, the bioavailability and physiological effects of these two forms of chromium appear to differ substantially.

Response to Comments: Separate RfCs have been generated for chromic acid mists and particulates of hexavalent chromium.

3. *Should the RfCs apply to both Cr(VI) and Cr(III) or only to Cr(VI)?*

A. Comment: The RfCs based on data from Lindberg and Hedenstierna and Glaser are appropriate only for Cr(VI).

4. *Are there any studies available that could be used to develop an RfC for trivalent chromium?*

A. Comment: The Agency has determined that an RfC for Cr(III) cannot be developed and I agree.

5. *The principal study (Mancuso, 1975) and the follow-up study (Mancuso, 1997) show the best dose-response relationship for total chromium, but animal data support a conclusion of carcinogenicity only for hexavalent chromium. Should the potency estimate address total chromium or hexavalent chromium?*

A. Comment: The potency estimate should be based on total chromium, but should note that the exposure is mixed.

Response to Comments: EPA will reevaluate the potency estimate for chromium based on the recent Mancuso update in the future.

B. Comment: The potency estimates should be based on hexavalent chromium.

Response to Comment: EPA will re-evaluate the potency estimate for chromium based on the recent Mancuso update in the future.

6. *There is a Canadian study that relates stomach cancer to gold mining following exposures to chromium. Does this study justify/support determination of an oral factor for chromium?*

A. Comment: I believe the Canadian study should not be used to determine an oral slope factor.

Response to Comment: The Canadian study has not been used to determine an oral slope factor.